

Safe, Effective, and Patient-Specific Glycaemic Control in Neonatal Intensive Care

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Nomenclature

<i>BG</i>	Blood glucose
<i>BM model</i>	Body mass model
<i>BW</i>	Birth weight
<i>cdf</i>	Cumulative distribution function
<i>CGM</i>	Continuous glucose monitor
<i>CNS</i>	Central nervous system
<i>CRIB2</i>	Clinical risk index for babies 2
<i>DOR</i>	Day of randomisation
<i>EBM</i>	Expressed breast milk
<i>ECV</i>	Extra-cellular volume
<i>EGP</i>	Endogenous glucose production
<i>ELBW</i>	Extremely low birth weight
<i>EN</i>	Enteral nutrition
<i>GA</i>	Gestational age
<i>GFR</i>	Glomerular filtration rate
<i>GI</i>	Glucose infusion
<i>GLUT</i>	Glucose transporters
<i>HC model</i>	Head circumference model
<i>HINT</i>	Hyperglycaemia and Insulin in Neonates Trial
<i>ICING model</i>	Intensive Care Insulin-Nutrition-Glucose model
<i>ICU</i>	Intensive care unit
<i>IGF-1</i>	Insulin-like growth factor 1
<i>IGUR</i>	Inter-uterine growth retardation
<i>IIT</i>	Intensive insulin therapy
<i>IQR</i>	Inter quartile range
<i>ISIG</i>	CGM measured current
<i>IV</i>	Intra-venous
<i>IVH</i>	intraventricular haemorrhage
<i>LBW</i>	Low birth weight
<i>NEC</i>	necrotising enterocolitis
<i>NICING model</i>	Neonatal Intensive Care Insulin-Nutrition-Glucose model
<i>NICU</i>	Neonatal intensive care unit
<i>NIRTURE</i>	Neonatal Insulin Replacement Therapy in Europe
<i>NZ</i>	New Zealand

<i>PICU</i>	Paediatric intensive care unit
<i>PN</i>	Parenteral nutrition
<i>PNA</i>	Post natal age
<i>PROM</i>	Premature rupture of membranes
<i>ROP</i>	Retinopathy of Prematurity
<i>SEM</i>	Standard error measurement
<i>SGA</i>	Small for gestational age
<i>SI</i>	Insulin sensitivity
<i>STAR</i>	Stochastic TARgeted
<i>STAR-GRYPHON protocol</i>	Stochastic TARgeted Glucose Regulation sYstem to Prevent Hyper- and hypO-glycaemia in Neonates protocol
<i>STAR-NICU protocol</i>	Stochastic TARgeted NICU protocol
<i>SUGAR-BABIES trial</i>	Randomised control trial of dextrose gel to treat hypoglycaemia in neonates
<i>TGC</i>	Tight glycaemic control
<i>TPN</i>	Total parenteral nutrition
<i>USA</i>	United States of America
<i>VLBW</i>	Very low birth weight

Abstract

In neonatal intensive care hyperglycaemia is a common complication of prematurity, and has been associated with worsened outcomes. While not as extensive as in adult and paediatric intensive care, there is still a great deal of debate and study surrounding the use of insulin therapy for glycaemic control in premature infants. It has been well established that insulin therapy allows greater glucose tolerance and weight gain.

However, the effect of insulin therapy on other clinical outcomes is not as well defined. In particular, a much higher incidence of hypoglycaemia is seen in many studies of insulin therapy in this cohort, which confounds results. Thus, the primary weakness of most studies in this area is that they seek to establish the effect of tight glycaemic control on outcomes without first developing a safe and effective protocol or tool for insulin therapy.

In neonatal intensive care there is no universal consensus on insulin therapy implementation, or well established threshold for hyperglycaemia and defined, outcome-based target range for control. When insulin therapy is used, many protocols are ad hoc, or utilise sliding scale based methods that fail to account for variability between patients, or changing patient condition over time. This situation is completely analogous to the adult intensive care situation.

Physiological model-based protocols offer great potential for allowing patient-specific care. Model-based tight glycaemic control methods have been shown to provide safe and effective control in adult intensive care units, and have demonstrated improved outcomes in treated patients. To date, only one such protocol exists in neonatal intensive care. The aim of this thesis is to improve on this protocol through more comprehensive modelling of glucose-insulin physiology, and optimisation of the clinical application of these methods.

Over the course of this thesis the NICING (Neonatal Intensive Care Insulin-Nutrition-Glucose) model is developed. This model is based on the best aspects of two previously clinically validated models in neonatal and adult intensive care, respectively. In particular, the resulting model is more physiologically descriptive than the previous neonatal glucose-insulin model.

A model of insulin secretion was developed specific to very premature infants using very unique data set collected from a recent study of glycaemic control in neonatal intensive care. In particular, C-peptide was used to provide quantitative estimates for insulin secretion, where previous work in the literature has only indirectly assessed it through peak plasma insulin concentration, or plasma insulin AUC. Results showed that insulin secretion was best modelled as a function of blood glucose concentration, and that it differed significantly between male and female very premature infants, a unique discovery in its own right.

Insulin clearance was examined, and different clearance pathways of insulin are modelled. These pathways include liver and kidney clearance, trans-capillary diffusion, and cellular degradation. In particular, kidney clearance is made more patient-specific, and interstitial fluid volume contraction following birth has also been incorporated into the model.

Endogenous glucose production was examined through extensive analysis of tracer studies in the literature for the very premature infant cohort. While endogenous glucose production varied significantly between patients and between studies, production was found to be suppressed with increasing glucose infusion over all studies, and suppressed with increasing blood glucose concentration over most of the studies. However, the major conclusion of the analysis was that variability outweighs the ability to derive a patient-specific model of endogenous glucose production. As a result, a simple constant for endogenous glucose production proved most effective in modelling and control.

Central nervous system uptake plays a significant role in non-insulin mediated glucose uptake. While relatively constant in adults, brain mass in premature infants is more variable than in adults, both between patients and over time. As a result central nervous system uptake is modelled as a constant proportional to brain mass, and two models for brain mass estimation were compared. Both head circumference and body mass based models resulted in similar model-based insulin sensitivity profiles, and thus similar control outcomes.

One significant inclusion in the new NICING model was a two compartment gut model. The two compartments of the new model include glucose appearance in the stomach and gut. In premature infants gastric emptying is slower than in term infants, so can be modelled with a half life of around 40 minutes ($d_1=0.017 \text{ min}^{-1}$), as opposed to around 20 minutes in term infants. CGM data showing the appearance of glucose from enteral feeds in term and premature infants was used to estimate the rate constant for glucose absorption from the gut

to the bloodstream. This absorption rate constant varied between infants and over time, and the median absorption constant was found to be $d_2=0.014 \text{ min}^{-1}$, which corresponds to an absorption half life of 50 minutes.

The new NICING model was used alongside stochastic forecasting methods in simulation to develop and optimise a STAR (Stochastic TARgeted) glycaemic control protocol for neonatal intensive care. This protocol used forecast ranges of likely insulin sensitivity outcomes to generate likely outcomes in blood glucose. Insulin doses were chosen such that these blood glucose outcome ranges overlapped a clinically specified target range. The resulting protocol, denoted the STAR-GRYPHON (Glucose Regulation sYstem to Prevent Hyper- and hypO-glycaemia in Premature Neonates) protocol, dosed insulin such that the 5th percentile of blood glucose predictions was within a lower limit tolerance of 4.0-4.4 mmol/L. Additional limits on the amount by which insulin can increase were applied. These limits are blood glucose dependent, and allow higher changes between doses at blood glucose levels greater than 9.0 mmol/L. They also limit the amount by which an insulin infusion can potentially increase to 0.01 U/kg/hr at blood glucose concentrations less than 6.0 mmol/L, thus adding additional safety from hypoglycaemia.

The STAR-GRYPHON protocol was implemented clinically. A total of 13 patients, spanning 16 glycaemic episodes, were treated under this protocol, and showed 76.6% of blood glucose within the clinically targeted range of 4.0-8.0 mmol/L. This performance is similar to the performance predicted in simulation of 76.9%. There were no incidences of blood glucose less than 3.5 mmol/L, and the incidence of hyperglycaemia was almost half that predicted in simulation. Overall performance is significantly better than retrospective and previous clinical results, and is better than any literature results to date, indicating that STAR-GRYPHON is safe and effective in clinical use.

Overall, a series of analyses have lead to a more physiologically descriptive model and optimised glycaemic control model and protocol for glycaemic control in very premature infants. Early results show significantly improved performance and safety over previous work and all the literature to date. Future work will follow up on its use in other New Zealand neonatal intensive care units (NICUs), and also NICU's in Europe.

Chapter 1 - Prematurity and Neonatal Glycaemia

The economics of healthcare are becoming increasingly difficult. Healthcare costs are increasing rapidly, driven in part by greater technological use and the increasing demand generated by higher critical illness/injury survival rates and aging populations (Bloomfield, 2003, Dorman and Pauline, 2007). Such economic trends are unsustainable, eroding the quality, access and equity of healthcare for all. In addition, the future of health is increasingly viewed as personalised, predictive, preventive and participatory (Hood and Friend, 2011), as well as needing to be more productive and economically affordable. In summary, better, more productive and less costly care is the necessary future of healthcare.

Disproportionate to both cohort and patient size, the neonatal intensive care cohort is one of the most vulnerable, and the most expensive, hospital cohorts. The costs associated with neonatal care increase with increasing prematurity, and are highest in premature infants born with a birth weight of less than 1000g birth weight (Kromhout et al., 1987). Despite higher mortality rates (Costeloe et al., 2012), costs associated with these infants are much higher due to increased incidence and severity of morbidities in surviving infants (Kromhout et al., 1987).

Hyperglycaemia, abnormally elevated blood glucose concentrations, is a common complication of prematurity, stress, and illness in neonatal intensive care. Hyperglycaemia has been associated with a range of common morbidities in neonatal intensive care. This chapter will first introduce prematurity and its incidence and outcomes. It will then describe the effect of prematurity on glucose and insulin metabolism, and its associated detrimental effects on outcomes. The purpose is to provide a case for research into the treatment of hyperglycaemia in this cohort, and a basis for the potential benefits of this therapy.

1.1 Prematurity

1.1.1 Definition

Premature infants are those born before the standard gestational period (in humans ~40 weeks) is completed. These infants are commonly classified according to their birth weight (BW) and/or gestational age (GA), as summarised in Table 1.1. While prematurity is commonly defined as infants born less than 37 weeks GA, this cohort is further divided into very premature (GA<31 weeks) and extremely premature (GA<27-28 weeks) infants. Alternatively, premature neonates may be categorised as low birth weight (LBW: BW<2,500g), very low birth weight (VLBW: BW<1,500g), extremely low birth weight (ELBW: BW<1,000g), or micropremie (BW<800g).

Table 1.1 Common classifications of prematurity

Classification by gestational age (GA)	Classification by birth weight (BW)
Premature : GA<37 weeks Very premature: GA<31 weeks Extremely premature: GA≤27 weeks	Low birth weight (LBW): BW<2,500g Very low birth weight (VLBW): BW<1,500g Extremely low birth weight (ELBW): BW<1,000g Micropremie: BW<800g

1.1.2 Epidemiology

Across Europe and most developed countries around 5-9% of all births are premature. This rate is higher in the USA with around 12-13% of all births being premature (Goldenberg et al., 2008, Tucker and McGuire, 2004). Table 1.2 provides a broad international set of rates of premature birth (Table 1.1). In New Zealand around 6.4% of all births are preterm (Beck et al., 2010). The incidence of premature birth before 32 weeks gestational age is around 1-2% (Tucker and McGuire, 2004).

Table 1.2: Worldwide rates of preterm birth. Adapted from: Beck et al. (2010)

Country	Preterm birth rate %
World Total	9.6
Africa	11.9%
Asia	9.1%
Europe	6.2%
North America	10.6%
Australia/New Zealand	6.4%

There are 3 main reasons for premature birth: 1) Maternal or foetal indications; 2) spontaneous labour; and 3) premature rupture of membranes (PROM). Maternal or foetal medical indications account for 30-35% of all preterm births (Goldenberg et al., 2008), and include cases such as preeclampsia, inter-uterine growth retardation (IGUR), small size for gestational age (SGA), foetal distress, and placental abruption, to name a few (Ananth and Vintzileos, 2006). Spontaneous labour accounts for 40-45% of preterm births, while PROM accounts for 35-35% of preterm birth in the USA (Goldenberg et al., 2008). Figure 1.1 provides a breakdown for the USA as an illustration.

In the USA, overall preterm birth rates are slowly rising (Figure 1.1). This rise is predominantly due to an increase in medically indicated preterm deliveries, although spontaneous preterm births have dropped slightly (Goldenberg et al., 2008). Other factors contributing to the increase in preterm births include increasing use of assisted reproduction techniques, changes in clinical practice, and misclassification of foetal loss/stillbirth/foetal death (Tucker and McGuire, 2004).

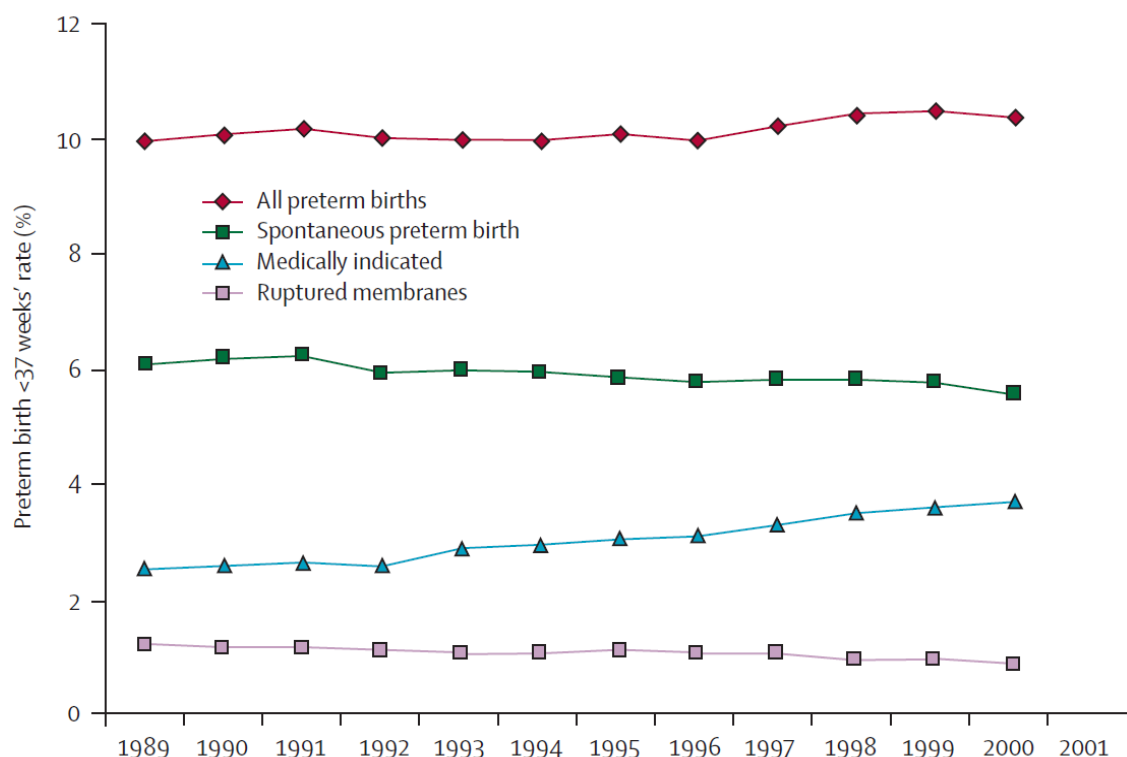


Figure 1.1: Preterm births in the USA by year and type. Source: Goldenberg et al. (2008).

Risk factors and causes of premature birth are hard to pinpoint, but include infection/inflammation, utero-placental ischemia or haemorrhage, uterine over distension, stress, or immunological mediated processes (Goldenberg et al., 2008). Prominent risk factors include: ethnicity, socio-economic factors, previous pregnancy history, multiple births (i.e. twins or triplets), and smoking (Goldenberg et al., 2008, Tucker and McGuire, 2004). In New Zealand, maternal smoking and multiple births have been identified as important risk factors (Wright et al., 1998).

1.1.3 Outcomes of Prematurity

It is estimated that 75-80% of all perinatal deaths occur in foetuses delivered prematurely, and that 40% of these deaths occur in infants born at less than 32 weeks gestation (Ananth and Vintzileos, 2006). Survival increases with gestational age and birth weight, as shown in Figure 1.2 and Table 1.3, with a gestational age of 22-23 weeks typically considered the limit for viability outside the womb. Over 22-27 weeks gestational age, the rates of survival increase significantly, and around 85% of preterm neonates born at 27 weeks gestational age survive the neonatal period.

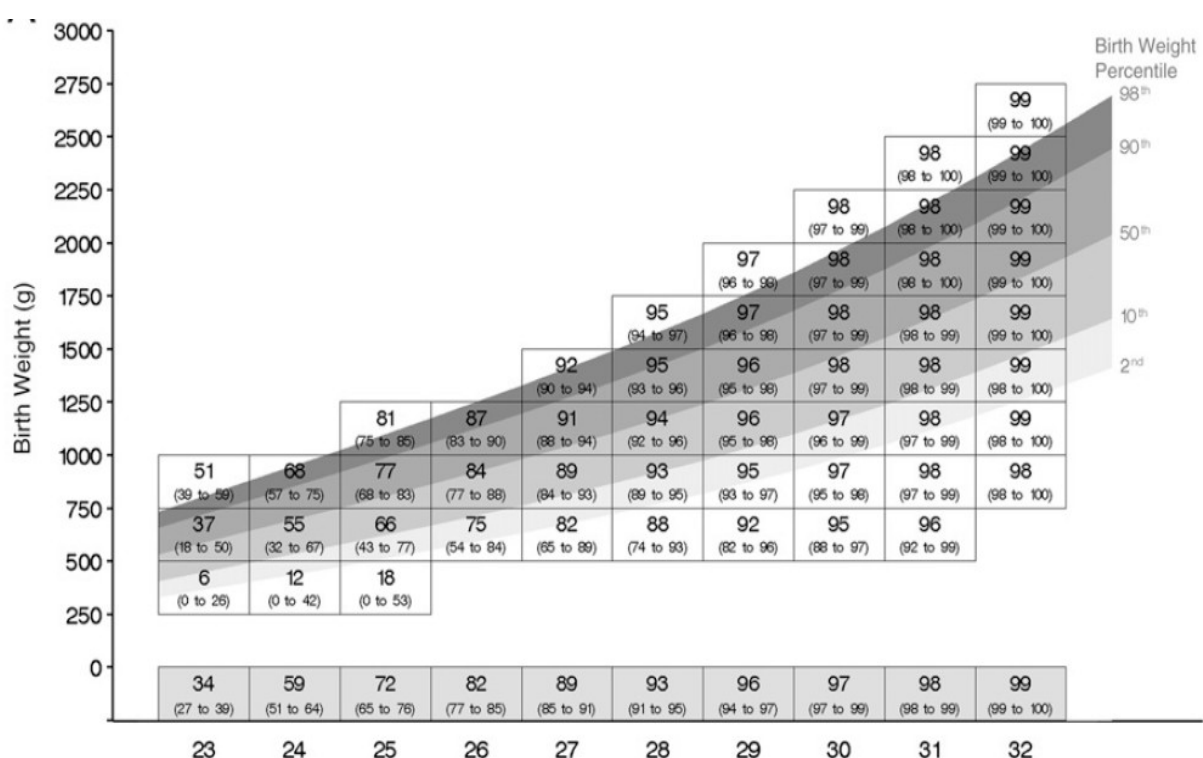


Figure 1.2: Population based model for prediction of survival rates in preterm infants.
Source : Manktelow et al. (2013).

Table 1.3: Survival outcomes in extremely preterm infants from several studies

	Survival rate (%)					
	22 weeks	23 weeks	24 weeks	25 weeks	26 weeks	27 weeks
Kyser et al. (2012)	33	58	87	85	-	-
Draper et al. (1999)	2	6	16	54	72	84
Manktelow et al. (2013)	-	27	52	65	77	85
Costeloe et al. (2012)	3	26	51	3	81	

In surviving infants, preterm birth is implicated in long term morbidities, developmental disabilities, and thus high healthcare costs (Ananth and Vintzileos, 2006). Further, because of increased survival, it is estimated that infants born with birth weight less than 1000g represent most of the patient days in the NICU (Farrag et al., 1997). Hence, these ELBW (Table 1.1) infants are of particular concern.

1.2 Neonatal Hyperglycaemia

1.2.1 Definitions and Thresholds

Hyperglycaemia is generally defined as abnormally elevated blood glucose (BG) concentration. There is no universally accepted quantitative description or threshold for clinical hyperglycaemia, or a best practice target range for glycaemic control. This lack of definitions was demonstrated in a recent survey of neonatal units in Australasia (Alsweiler et al., 2007), where there was significant variation in the range of basal BG levels targeted. Figure 1.3 summarises these results, and clearly shows a wide range of expectations.

While clinical hyperglycaemia is commonly defined as BG>10 mmol/L (180 mg/dL), in the literature this threshold is often significantly lower at 6.9-8.3 mmol/L (125-150 mg/dL) (Cowett et al., 1979, Hall et al., 2004, Hays et al., 2006, Louik et al., 1985, Vaucher et al., 1982). Current working definitions for hyperglycaemia (BG> 7-8 mmol/L) are based on statistical distributions of BG levels in term infants (Hey, 2005). However, these statistical definitions may describe what is “normally” found, but not necessarily what is clinically desirable in this cohort (Hey, 2005).

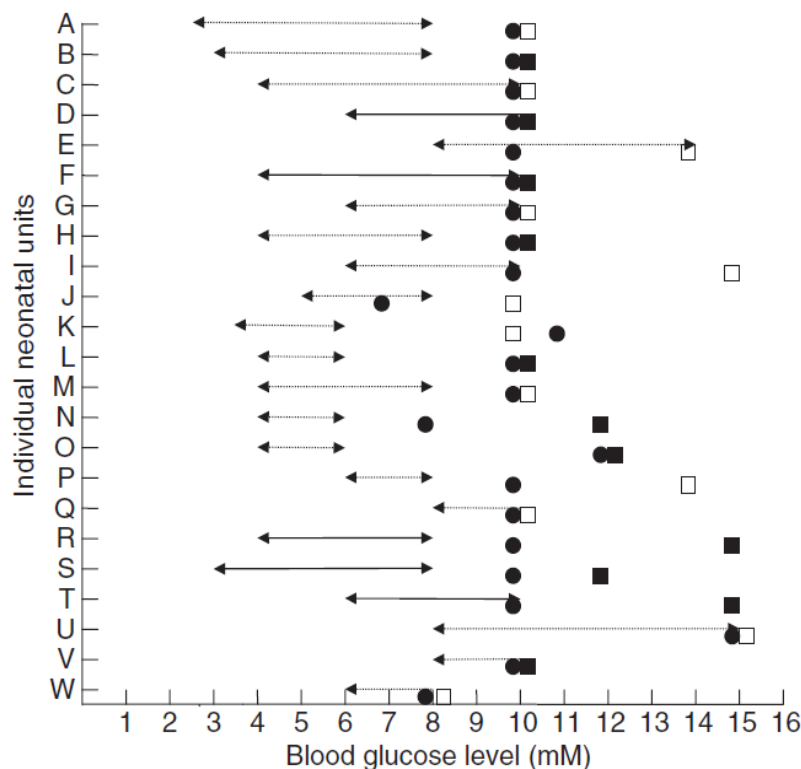


Figure 1.3: Definition of hyperglycaemia, criteria for starting insulin, and blood glucose level (mM is mmol/L) range targeted for babies on insulin in neonatal units in Australasia. Circles show definition of hyperglycaemia, open/closed squares show the threshold for starting insulin therapy (without/with additional glycosuria thresholds). Arrows define the target blood glucose range for a unit. Source Alsweiler et al. (2007).

For example, Figure 1.4 shows the distribution of BG levels in preterm infants of gestational age 32 weeks or less. While the 50th percentile is roughly “normal,” a high percentage of BG levels were hypoglycaemic (BG<2.5 mmol/L), and also hyperglycaemic (BG>7-8 mmol/L), by statistical definition. In addition, no studies have attempted to define hyperglycaemia in the ELBW preterm infant (BW<1000g) (Pildes and Pyati, 1986). Thus there is a lack of clear definition of what are acceptable levels of BG in these cohorts.

In his review, Hey suggests a pragmatic working definition of hyperglycaemia is BG>12mmol/L (Hey, 2005). However, as shown in Figure 1.3, insulin treatment is often initiated at much lower BG levels. In the Christchurch Women’s Hospital NICU, the clinical centre for this thesis, hyperglycaemia is clinically defined as BG>10.0 mmol/L, and insulin therapy is initiated with two consecutive BG>10.0 mmol/L.

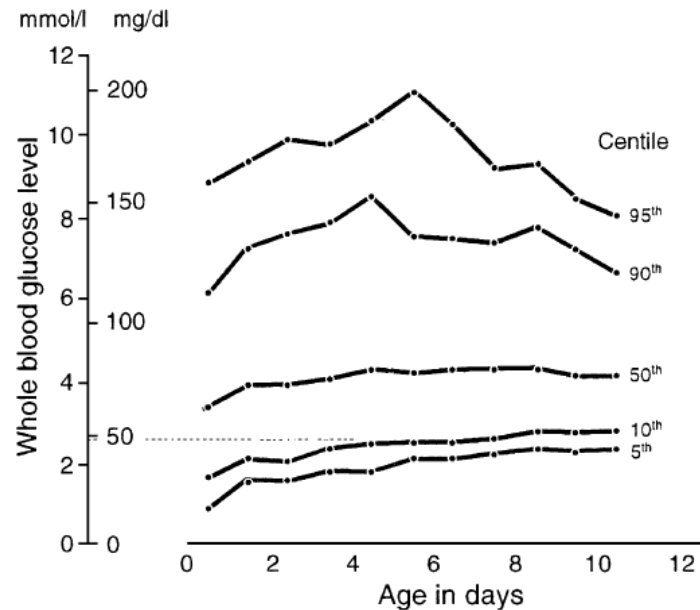


Figure 1.4: Laboratory estimates of the whole blood glucose from a prospective study of babies born <32 weeks in England. Source: Hey (2005).

1.2.2 Prevalence of Hyperglycaemia

Hyperglycaemia is a common complication of prematurity, particularly with lower gestational age and/or lower birth weight infants (Lilien et al., 1979, Louik et al., 1985). Table 1.4 reflects this increasing prevalence in lower birth weight infants across a number of studies. Differences in the definition of hyperglycaemia and weight ranges between studies make overall incidence difficult to determine. However, it can be reasonably stated that around 30-50% of infants with birth weight less than 1.5 kg are likely to have a least one BG>8.3 mmol/L (150 mg/dL). Hyperglycaemia is more common in infants receiving more than 8 mg/kg/min of glucose infusion (Brans et al., 1974, Cowett et al., 1979, Pildes and Pyati, 1986), whereas the term infant is better able to tolerate much greater glucose delivery (Brans et al., 1974).

1.3 Background Physiology: Glucose-Insulin Metabolism.

Glucose is a six carbon sugar that is a key fuel source in the body. When food is ingested, complex carbohydrates and sugars are broken down into glucose, which is the fundamental building block of carbohydrate molecules. The glucose is then absorbed into the blood stream, and is either utilised for energy or stored as fat or glycogen for later utilisation.

Table 1.4: Prevalence of hyperglycaemia over several studies

Study	Cohort	Incidence
Dweck and Cassady (1974)	Birth weight<1.1kg	<ul style="list-style-type: none"> • 85% had episode(s) of BG> 6.9 mmol/L (125 mg/dL) • 72% had episode(s) of BG> 16.7 mmol/L (300 mg/dL)
Cowett et al. (1979)	AGA preterm infants with BW<1.5kg	<ul style="list-style-type: none"> • 0% of infants receiving ~8 mg/kg/had BG>8.3 mmol/L (150 mg/dL) • 50% of infants receiving ~11 mg/kg/min had maximum BG> 8.3 mmol/L (150 mg/dL), and 50% had maximum BG> 11.1 mmol/L (200 mg/dL) • 100% of infants receiving ~14 mg/kg/min had maximum BG> 8.3 mmol/L (150 mg/dL), and 50% had maximum BG> 11.1 mmol/L (200 mg/dL)
Louik et al. (1985)	Preterm infants	<ul style="list-style-type: none"> • BW<1kg: 29% had episode(s) of BG>8.3 mmol/L (150 mg/dL) • 1kg<BW<1.5kg: 8.6% had episode(s) of BG>8.3 mmol/L (150 mg/dL)
Heimann et al. (2007)	BW<1.5kg	<ul style="list-style-type: none"> • 49.6% had 1-3 BG measures > 8.3 mmol/L (150 mg/dL) • 19.9% had more than 4 BG measures > 8.3 mmol/L (150 mg/dL)
Hays et al. (2006)	BW<1kg	<ul style="list-style-type: none"> • From days 2-7, 57% had BG> 8.3 (150 mg/dL), and 32% had BG> 13.9 mmol/L (250 mg/dL) • Of ELBW infants, 42% had BG> 8.3 mmol/L (150 mg/dL) consistently • Of all infants, 25% had consistent BG> 11.1 mmol/L (200mg/dL), and 16% had consistent BG> 13.9 mmol/L (250 mg/dL)
Hall et al. (2004)	Infants with necrotising enterocolitis BW: 1.31 ± 0.09kg SEM	<ul style="list-style-type: none"> • 69% of infants had episode(s) of BG> 8 mmol/L
Zarif et al. (1976)	BW<2kg	<ul style="list-style-type: none"> • BG> 6.9 mmol/L (125 mg/dL) was observed in 32 of 75 infants (43%)
Brans et al. (1974)	Term and premature (BW<1.5KG)	<ul style="list-style-type: none"> • BG> 6.9 mmol/L (125 mg/dL) was observed in all premature infants (n=11). No hyperglycaemia was seen in term infants at all feed rates.

In contrast to muscle cells, which have a local store of glycogen, central nervous and neuronal tissues require a constant supply of free glucose (Tirone and Brunicardi, 2001). The brain is the single largest consumer of glucose in the adult body, accounting for approximately 50% of the body's glucose use, while 25% is used by the liver, and the remainder by insulin-dependent tissues, such muscle and adipose tissue (Triplitt, 2012). Thus, the brain and other neuronal tissues are heavily reliant on a constant supply of glucose

delivered via the blood stream as their main fuel source. Accordingly, blood glucose concentrations are regulated to a tight normal range, and concentrations outside this range either deprive cells of fuel or have patho-physiological effects. Regulation of blood glucose involves the complex interaction of a number of blood species, including glucose, insulin, glucagon, and a number of hormones, shown schematically in Figure 1.5.

1.3.1 Glucose Production and Clearance

Within the body, glucose can be produced from fat breakdown, from proteins via gluconeogenesis, and from glycogen via glycogenolysis. Gluconeogenesis occurs primarily in the liver, although after prolonged fasting and sustained acidosis it can also occur in the kidneys (Tirone and Brunicardi, 2001). Gluconeogenesis is regulated by pancreatic hormones, intracellular substrate levels, free fatty acid levels, and counter regulatory hormones. In addition, it can also be regulated by the central nervous system, whereupon sympathetic and parasympathetic nerves stimulate glucose production in the liver in response to hypoglycaemia in the brain (Tirone and Brunicardi, 2001).

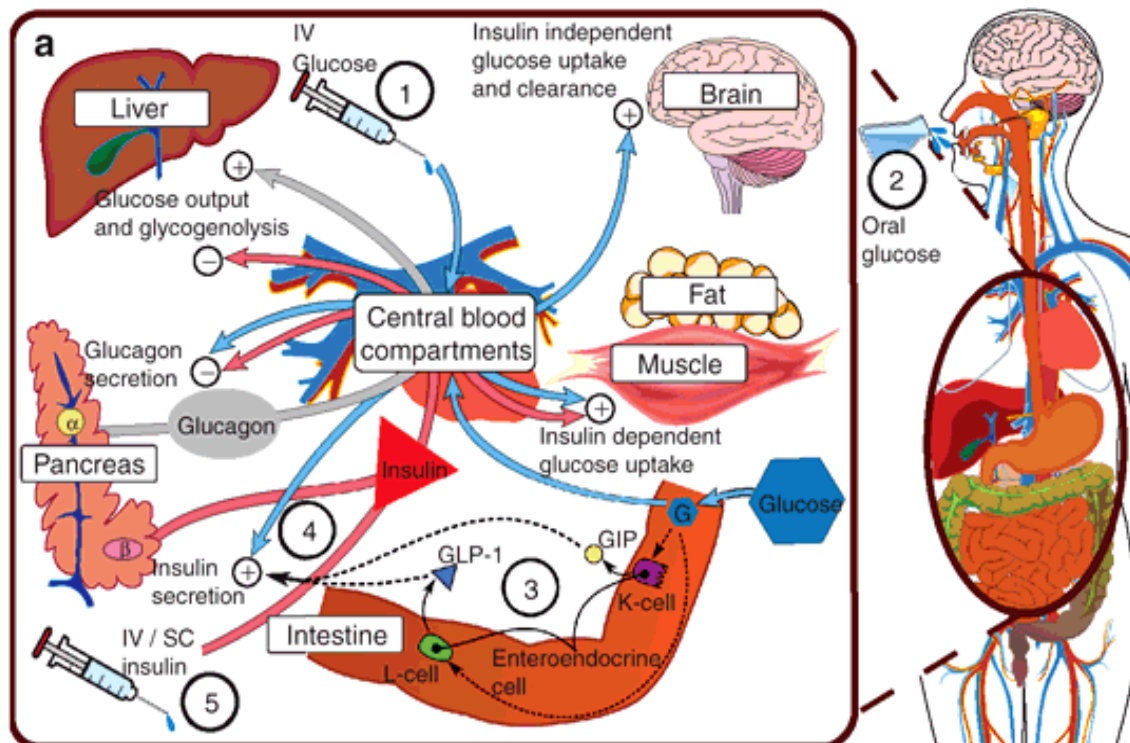


Figure 1.5: The glucose insulin regulation system. Blue, red, and grey arrows represent glucose, insulin, and glucagon respectively. Plus (+) and minus (-) signs are used to described the effect of glucose, insulin, and glucagon and metabolic process. Source: Schaller et al. (2013).

Gluconeogenesis is supplemented by glycogenolysis, which is the process that converts glycogen stores to glucose for use. In an adult, glycogen stores in the body can be used for approximately 8 hours before they run out (Tirone and Brunicardi, 2001). Following a 10 hour fast, gluconeogenesis makes up approximately 30% of total hepatic glucose production, while the rest is produced via glycogenolysis (Tirone and Brunicardi, 2001).

Counter-regulatory hormones, such as cortisol, growth hormones, thyroid hormones, and catecholamines like adrenalin, epinephrine, and norepinephrine, and angiotensin II, can stimulate hepatic glucose production. For example, epinephrine stimulates glycogenolysis, cortisol stimulates gluconeogenesis, and norepinephrine and angiotensin indirectly promote glycogenolysis via intracellular signalling (Tirone and Brunicardi, 2001). Thus, these hormones, often seen as part of any stress response, can also stimulate energy creation as part of the "fight or flight" reflex.

Glucose uptake by cells is facilitated by glucose transporters (GLUTs). These transporters provide an aqueous pore through which glucose can pass through the hydrophobic cell membrane. Once glucose enters a cell it is used either as a fuel source or is stored. It cannot leave the cell after entering. There are 5 main types of GLUT transporter, varying by tissue location (Tirone and Brunicardi, 2001, Triplitt, 2012). These are summarised in Table 1.5. Importantly, the GLUT4 glucose transporter is the insulin dependent transporter that enables the muscle/adipose cell to take up glucose via insulin-mediated exchange.

1.3.2 Insulin Secretion and Clearance

Insulin is an important agent in the regulation of blood glucose. Insulin mediates the uptake of glucose in muscle and adipose tissues by increasing the number of active GLUT 4 transporters in the cell membrane (Tirone and Brunicardi, 2001). Insulin is produced by Beta cells in the Islets of Langerhans in the pancreas, and is secreted into the portal vein in response to elevated blood glucose levels, such as following glucose ingestion (Tirone and Brunicardi, 2001, Triplitt, 2012). There is a first pass hepatic extraction of this insulin of ~30-80% (Ferrannini and Cobelli, 1987, Meier et al., 2005, Toffolo et al., 2006) before the remainder is transported around the body. Elevated plasma insulin concentrations restrict lipolysis and proteolysis, reducing glucose precursor availability to the liver, and thus, in normal function, inhibiting hepatic glucose production (Tirone and Brunicardi, 2001).

Table 1.5: Prominent glucose transporters in the human body

Glucose Transporter	Tissues found in	Insulin dependency	Action
GLUT1	All cells	Independent	Particularly responsible for glucose transport across blood-brain barrier
GLUT2	Primarily pancreas and liver	Independent	Not easily saturated, therefore allows Beta cells in Pancreas to be sensitive to changes in glucose concentrations
GLUT3	Neurons, intestine, testicles, kidneys	Independent	Allows uni-directional co-transport of sodium and glucose in small intestine and proximal tubules of kidney. Can transport glucose against glucose concentration gradient when it is along a sodium concentration gradient.
GLUT4	Muscle and Adipose tissue	Dependent	Insulin causes translocation of GLUT4 from intracellular vesicles to the cell membrane
GLUT5	Jejununal brush border (intestines)	Independent	Transports Fructose

The liver and the kidneys are the primary sites of insulin clearance in the body, accounting for the majority of peripheral insulin clearance. Liver clearance of insulin increases with nutrient uptake via the gut, due to hormone signalling. A glucose infusion delivered directly into the portal vein does not have this effect. Liver clearance of insulin is also a receptor-binding process, so while at elevated plasma insulin concentrations overall insulin clearance may be higher, the total fractional extraction is often lower, (Duckworth et al., 1998).

The kidneys clear approximately 50% of peripheral circulating insulin, and are also responsible for the clearance of around 50% of pro-insulin, an insulin precursor, and 70% of C-peptide, a molecule secreted in equal molar quantities with insulin due to the cleavage of pro-insulin into insulin and C-peptide. Insulin is cleared in the kidneys via glomerular filtration and absorption from the peritubular capillaries (Rabkin et al., 1984). Cleared insulin is degraded, so very little insulin is eventually secreted into urine (Duckworth et al., 1998).

1.3.3 Glucagon's Regulatory Effect

Glucagon is another regulatory hormone secreted by the Alpha cells in the Islets of Langerhans in the pancreas. Elevated plasma insulin levels, and/or low blood glucose levels,

stimulate glucagon secretion by the pancreas and mobilise glucogenetic precursors. Elevated glucagon concentrations in the portal vein stimulate both gluconeogenesis and glycogenolysis in the liver. Glucagon's effect on gluconeogenesis is persistent, whereas its effect on glycogenolysis is more short lived (Tirone and Brunicardi, 2001). In turn, the increased hepatic glucose production raises blood glucose levels, which suppresses the secretion of glucagon (Tirone and Brunicardi, 2001). Glucagon is thus an important and powerful hormone that is used to raise blood glucose levels and ensure the brain and other organs receive enough glucose to meet their demands.

1.3.4 Glycaemic Levels and Hormones

Overall, blood glucose levels are tightly regulated by a range of hormone actions. Blood glucose levels are raised by gluconeogenesis and glycogenolysis, instigated by glucagon or counter-regulatory hormones, such as cortisol, growth hormones, thyroid hormones or catecholamines. Blood glucose levels are lowered primarily by insulin mediated uptake by the liver and periphery. In normal function, these hormones act in concert to maintain or restore normal glycaemic levels or glucose homeostasis.

1.4 Neonatal Glycaemia

1.4.1 Foetal Glycaemic Development

While in the womb, foetal glucose homeostasis is dependent on placental glucose transfer, where the foetal glucose pool is in equilibrium with the mother's, and foetal endogenous glucose production is minimal or non-existent (van Goudoever et al., 1993). During a normal term birth the supply of glucose is interrupted, and the infant's body mounts an endocrine stress response. Catecholamines concentrations, such as adrenaline and epinephrine, cortisol, and glucagon, increase, and plasma insulin concentrations decrease. The low plasma insulin to glucagon ratio then stimulates both glycogenolysis and gluconeogenesis, and lipolysis and muscle protein breakdown provides substrates for these processes (van Goudoever et al., 1993). Glucose uptake in all cells is primarily due to the presence of GLUT1 transporters in the foetus (Cowett and Farrag, 2004). After birth, GLUT1 decreases and the amounts of other GLUT transporters increase (Cowett and Farrag, 2004).

However, in the case of the premature infant, the glucose regulation system is under-developed. In the womb, the foetal pancreas appears to be the major source of insulin (Adam

et al., 1969b), and it been shown to secrete insulin in response to glucose or amino acids from the 20th week of gestation (van Goudoever et al., 1993). Overall foetal plasma insulin levels are higher than in the adult case, but the action of this insulin is modulated by glucocorticoids and is inactive until sometime later in the second trimester of pregnancy (weeks 13-28) (van Goudoever et al., 1993). One study suggests that bi-phasic insulin secretion in response to nutrient release does not begin to mature until after birth (Otonkoski et al., 1988). Thus, premature infants have reduced capacity to secrete and utilise insulin to lower blood glucose, in comparison to their term counterparts.

Glycogen synthesis starts in the second trimester, and increases slowly until the 36th week of gestation. Glycogen is not stored until the 27th week of gestation (van Goudoever et al., 1993). Thus, if an infant is extremely premature (<27 weeks gestation) it lacks the glycogen stores of adults and older babies that are used to provide energy. These infants typically require exogenous intra-venous (IV) glucose infusion soon after birth to prevent hypoglycaemia. In addition, premature infants have also been shown to have inadequate glucagon secretion at low blood glucose levels, and thus have low hepatic glucose production, even if substrates are available (Hume et al., 2005). Hence, these infants have inadequate and dysfunctional regulation with respect to the ability to raise low blood glucose levels.

Similarly, in the presence of an exogenous glucose infusion persistent endogenous glucose production was observed in new born infants, particularly in the premature infant group (Cowett et al., 1983). This result is in to contrast with the normal adult case, where endogenous glucose production is suppressed with high exogenous glucose infusion (Wolfe et al., 1979). Thus, equally, there is a dysfunctional regulation response to higher blood glucose levels and availability.

1.4.2 Causes of Neonatal Hyperglycaemia

Defective beta-cell processing of proinsulin

Pro-insulin is a polypeptide chain that is a precursor to insulin and C-peptide, and is 10 times less active than insulin (van Goudoever et al., 1993). It has been shown that the pancreas of a premature neonate is sensitive to changes in blood glucose, and will secrete pro-insulin in response to elevated blood glucose levels and decrease secretion in response to an insulin-infusion induced drop in blood glucose. However, insulin levels were not significantly

different than in euglycaemic (normal blood glucose) premature infants, suggesting defective processing of the proinsulin to insulin (Mitanezh-Mokhtari et al., 2004), which enables rising glucose.

Relative insulin resistance

Elevated plasma insulin and C-peptide concentrations of preterm infants in comparison to term infants has been used as evidence to suggest that premature infants are more resistant to insulin. In addition, similar plasma insulin concentrations in hyperglycaemic infants when compared to euglycaemic infants, and subsequent treatment with exogenous insulin to bring about euglycaemia, suggests that hyperglycaemic premature infants are more resistant than their euglycaemic counterparts (Mitanezh-Mokhtari et al., 2004). Thus, these infants are less able to use insulin to lower blood glucose levels than normal, term infants.

No suppression of endogenous glucose production

Several studies show that endogenous glucose production in the term infant persists, despite exogenous glucose infusions around basal rates (4-6 mg/kg/min) (Cowett et al., 1983, Sunehag et al., 1994b). Only one study has observed suppression of endogenous glucose production with intravenous glucose (Hertz et al., 1993). This lack of suppression contrasts the normal adult case in which endogenous glucose production is suppressed with high exogenous glucose infusion (Wolfe et al., 1979). Equally, glucose production has also been shown to persist in the presence of exogenous insulin infusion (Farrag et al., 1997), where the normal response would be suppression. Hence, unsuppressed endogenous glucose production enables rising blood glucose.

Glucose Transporters (GLUTs)

In new-borns and premature infants the concentrations of the GLUT1 transporter are relatively high, while other transporters, including GLUT2 are low (Mitanezh, 2007). The lower number of GLUT2 transporters (see Table 1.5) could mean that pancreatic beta cells are not as sensitive to changes in blood glucose as in adults, thus reducing the degree to which the liver can respond to increased glucose and insulin concentrations (Farrag and Cowett, 2000). In addition, diminished GLUT4 transporters would limit the amount of insulin-mediated glucose uptake by muscle and adipose tissue (Farrag and Cowett, 2000). All these effects reduce the ability to lower elevated blood glucose levels.

Infection

Hyperglycaemia has also been identified as a predictor for septicaemia in this cohort (Fanaroff et al., 1998). In a trial of glycaemic control, increased sepsis was seen in the control group, and was identified as a contributor to glucose intolerance (Collins et al., 1991). In turn, infection is associated with increased neurologic morbidities (Van Kempen et al., 2005a). However, whether hyperglycaemia is a risk factor or a result of infection is debated, with both likely to be true in reality (Bistrian, 2001). Metabolically, infection reduces the ability of insulin to act.

Stress

Stress leads to counter-regulatory hormone secretion. In some studies stress was shown to be a significant factor responsible for hyperglycaemia in a study of VLBW premature infants (Table 1.1) (Lilien et al., 1979, Louik et al., 1985). In a study of term infants during surgery and anaesthesia, hyperglycaemia was a commonly observed complication (Anand et al., 1987, Anand et al., 1988, Srinivasan et al., 1986).

The pathophysiology of stress hyperglycaemia is summarised in Figure 1.6. Stress can cause increased levels of cortisol, glucagon and catecholamines in the blood, which increase endogenous glucose production (Tirone and Brunicardi, 2001). Increased levels of these molecules are associated with conditions such as surgery and anaesthesia (Anand et al., 1987), respiratory distress, cardiac failure, sepsis, and necrotising enterocolitis (Farrag and Cowett, 2000). In adults, they are also associated with suppressed insulin production and action (McCowen et al., 2001). All these outcomes enable hyperglycaemia.

Drug therapy

Hyperglycaemia may also be drug related. For example steroids, methylxanthines (theophylline), or dopamine (Farrag and Cowett, 2000) can raise endogenous glucose production, and/or antagonise the secretion and action of insulin to lower insulin. This effect has also been observed in critically ill adults (Pretty et al., 2012). Thus, hyperglycaemia may be iatrogenically induced by other necessary therapies.

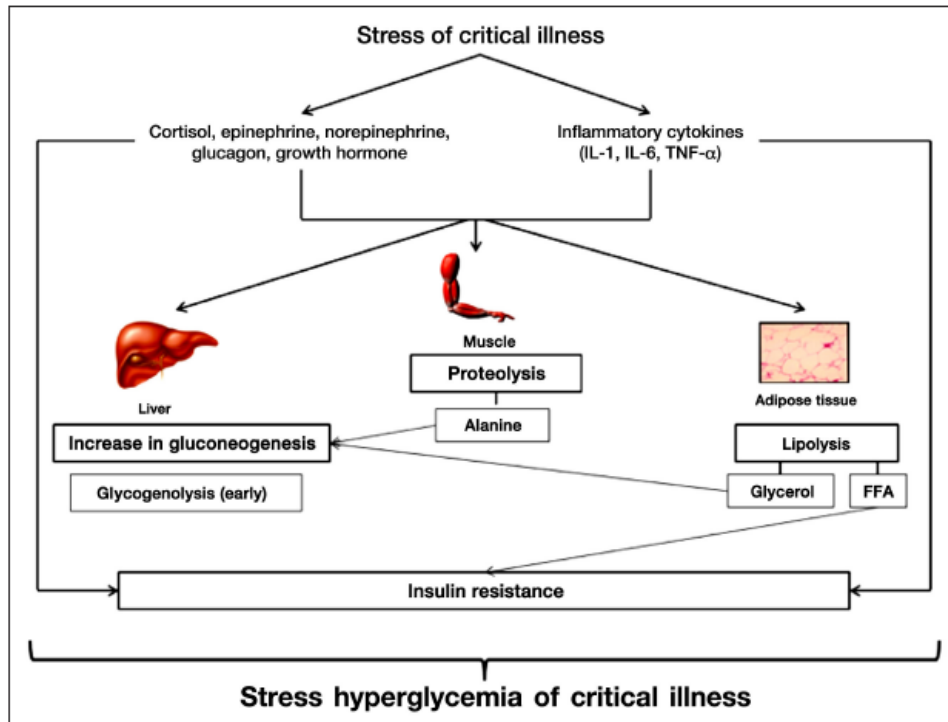


Figure 1.6: Pathophysiology of stress hyperglycaemia in critical illness. Source: Reitano et al. (1978).

Exogenous glucose infusion

The incidence of hyperglycaemia has been shown to increase with higher glucose infusions (Cowett et al., 1979, Dweck and Cassady, 1974, Farrag and Cowett, 2000, Louik et al., 1985). Cowett et al. (1979) found that infants receiving ~8mg/kg/min of glucose had blood glucose and plasma insulin levels similar to baseline steady state measures. However, in the groups receiving ~11-12 mg/kg/min and ~14 mg/kg/min there were incidences of hyperglycaemia (BG>8.3 mmol/L (150 mg/dL)) and glucosuria (trace or greater glucose in the urine) in half and all of the infants respectively. In comparison, term infants are better able to tolerate higher than normal glucose infusions (Brans et al., 1974). Hence, these infusions, coupled with other factors, may be another iatrogenic source of hyperglycaemia.

1.4.3 The Effect of Hyperglycaemia on Outcomes in Critical Care

In adult and paediatric intensive care there is a wealth of literature relating to the effect of hyperglycaemia on outcomes and the use of insulin to treat it. Hyperglycaemia in intensive care patients has been associated with increased mortality (Capes et al., 2000, Holm et al., 2004, Krinsley, 2006, Sperry et al., 2007), organ failure (Chase et al., 2010b, Sperry et al.,

2007, Van den Berghe et al., 2001), and risk of congestive heart failure or cardiogenic shock (Capes et al., 2000). In the paediatric ICU (PICU) hyperglycaemia is associated with increased mortality (Baumeister et al., 2001, Cser and Milner, 1975, Mitanchez, 2007), high wound infection rates (Van Kempen et al., 2003b), increased mechanical ventilation (Grasso et al., 1973), increased length of stay (Grasso et al., 1973) and total length of hospital stay in abdominal surgery patients (Wu et al., 2013). In trauma patients it has been associated with increased mortality (Wahl et al., 2008), length of hospital stay (Bochicchio et al., 2007, Bochicchio et al., 2005, Yendamuri et al., 2003), ventilator dependence (Bochicchio et al., 2007, Bochicchio et al., 2005, Sung et al., 2005), and infection (Bochicchio et al., 2007, Bochicchio et al., 2005, Laird et al., 2004, Sung et al., 2005). It has also been suggested that the relationship between hyperglycaemia and mortality/morbidity may be stronger in trauma patients as opposed to those in surgical intensive care (Vogelzang et al., 2006).

In addition, there is debate as to whether hyperglycaemia is a cause or result of infection, or both (Bistrian, 2001). In the adult and paediatric patient, elevated blood glucose levels have been associated with sepsis and septic shock (Branco et al., 2005). Elevated blood glucose levels have also been associated with increased infection related mortality (Yendamuri et al., 2003).

Although far from as extensive as the adult literature in this area, a number of studies in preterm neonates have been conducted to assess the effect of hyperglycaemia on clinical outcomes in this cohort. Overall, these studies indicate that hyperglycaemia is associated with increased morbidity and mortality. The definitions of hyperglycaemia differ across articles, but overall conclusions tend to be consistent. In particular:

Mortality: Hyperglycaemia has been associated with increased mortality (Alaadeen et al., 2006, Hall et al., 2004, Hays et al., 2006). It has been shown that this association is particularly strong in the case of severe early (Kao et al., 2006) and persistent or repeated hyperglycaemia (Heimann et al., 2007, Kao et al., 2006).

Hospital length of stay: Although one study saw no association of hyperglycaemia with hospital length of stay (Kao et al., 2006), the majority of the literature suggests that the two are linked (Alaadeen et al., 2006, Hall et al., 2004, Hays et al., 2006), particularly in the case of persistent or prolonged hyperglycaemia (Hays et al., 2006).

Ventilator dependence: It has been suggested that hyperglycaemia is associated with increased duration of mechanical ventilation (Alaedein et al., 2006), but another study has not seen this result (Kao et al., 2006).

Intraventricular Haemorrhage: Increased incidence of severe intraventricular haemorrhage is associated with hyperglycaemia (Hays et al., 2006).

Retinopathy of prematurity: Hyperglycaemia is associated with increased incidence of retinopathy of prematurity (ROP) (Blanco et al., 2006, Ertl et al., 2006, Garg et al., 2003).

1.4.4 Hypoglycaemia

Hypoglycaemia, or low blood glucose levels, can also occur frequently in premature infants, even in the absence of insulin therapy (Sexson, 1984). By definition, hypoglycaemia results in significantly restricted availability of metabolic fuel for cell function and growth, as well as to the brain. Thus, it is also associated with adverse neurological outcomes in neonates (Koh et al., 1988a, Lucas et al., 1988).

Definitions of hypoglycaemia and its clinical significance have varied historically, and between studies (Cornblath et al., 2000, Koh et al., 1988a, Koh et al., 1988b, Sexson, 1984). Across medical textbooks thresholds range from 1.7 – 2.5 mmol/L, while clinically it can vary from 1.0 – 4.0 mmol/L, with a modal average of 2.0 mmol/L in one review (Koh et al., 1988b). Historically, clinically significant hypoglycaemia had to fulfil a threefold criterion: 1) clinical manifestations; which are 2) coincident with low blood glucose measurements; and 3) with clinical symptoms resolving within minutes when euglycaemia is re-established. However, this definition excludes infants with extremely low blood glucose levels that were asymptomatic (Cornblath et al., 2000).

Symptomatic hypoglycaemia may manifest in the infant as tremors, sweating, sudden apathy or limpness, cyanosis and apnea, high-pitched cry, convulsions, coma, or cardiovascular collapse (Cornblath et al., 2000, Pildes and Pyati, 1986). It is generally regarded to be more serious than asymptomatic hypoglycaemia (Cornblath and Schwartz, 1976). Controversy remains as to the effect of asymptomatic hypoglycaemia on the brain (Cornblath et al., 1990, Koh et al., 1988b). Treatment of hypoglycaemia can include intravenous boluses of dextrose to induce euglycaemia. A recent study in New Zealand has investigated the use of dextrose gel applied to the gums to prevent transient hypoglycaemia in the first few days of life (Harris

et al., 2013), and long term follow up assessing cognitive and developmental impacts is currently in progress (Harris et al., 2014).

1.5 Summary

Worldwide, around 9.6% of all births are premature and 1-2% of all births are very premature. Increasing prematurity is associated with increased severity of illness, and thus increased risk of mortality and increased burden of care. Higher degrees of prematurity are also associated with increased incidence of hyperglycaemia, which can affect up to 70% of extremely premature infants in neonatal intensive care. In turn, it is well established that hyperglycaemia is associated with increased risk of mortality and morbidities, and, in particular, a higher risk of infection. Controversy remains as to whether hyperglycaemia is a marker for, or a cause of, illness and injury, with the likely case that both are true.

Much research is being carried out in neonatal and wider intensive care settings to investigate the benefits of insulin therapy and glycaemic control. However, hypoglycaemia is also dangerous, can commonly occur with insulin therapy, and is associated with adverse neurologic outcomes. If the incidence of hyperglycaemia can be reduced, without increasing the incidence of hypoglycaemia, and it translates to a reduction in the incidence of morbidities, then not only is long term patient outlook improved, but the burden of the cost of care may also be reduced.

Chapter 2: The Problem of Variability: Glycaemic Control in Neonatal Intensive Care

Hyperglycaemia is an extremely common complication of prematurity (Brans et al., 1974, Dweck and Cassady, 1974, Hall et al., 2004, Louik et al., 1985, Zarif et al., 1976) and has been associated with increased mortality and morbidity (Alaedein et al., 2006, Garg et al., 2003, Hall et al., 2004, Hays et al., 2006, Hey, 2005, Kao et al., 2006). Thus, it is not surprising that increasingly blood glucose (BG) control is becoming a concern in intensive care contexts in general. In particular, the body of literature around glycaemic control in premature infants is increasing.

Insulin facilitates the transport of glucose into cells, and is widely used to treat hyperglycaemia in diabetic and in adult intensive care cohorts. Its use in critically ill adults for tight glycaemic control is endorsed by many organisations (Wiener et al., 2008), including the Surviving Sepsis Campaign (Dellinger et al., 2004), American Association of Clinical Endocrinologists (AACE, 2002), and the American Diabetes Association (Grasso et al., 1990). In paediatric care, the use of insulin to treat hyperglycaemia has been supported by the American Academy of Paediatrics since 1985 (American Academy of Pediatrics Committee on Nutrition, 1985).

Ideally, a glycaemic control protocol lowers BG to a euglycaemic range without driving BG concentrations dangerously low. To meet this goal, it must account for changes in patient condition over time, as glucose-insulin metabolism is affected by prematurity (Louik et al., 1985), stress (Lilien et al., 1979, Louik et al., 1985), and drug therapy (Farrag and Cowett, 2000), among many factors. This chapter evaluates current clinical practice and its limitations, as well as reviewing the effect of insulin therapy to treat hyperglycaemia on clinical outcomes. Model based methods are presented as a potentially more effective tool for managing patient variability, and thus for glycaemic control, than conventional therapy approaches.

2.1 Insulin Therapy and Outcomes in Intensive Care

While the association of hyperglycaemia with increased mortality and morbidities is increasingly well established, the use of insulin therapy to treat this condition and its effect on outcomes is less clear. As there is a larger body of literature surrounding the use of insulin therapy in adult and paediatric care, its effect on outcomes in these cohorts will be briefly reviewed first, as the main risks and elements are defined. It is then followed by a review of insulin therapy in neonatal intensive care.

2.1.1 Adult and Paediatric Intensive Care

In a few early studies, insulin therapy to maintain euglycaemia was shown to either leave unchanged (Van den Berghe et al., 2006b) or reduce mortality rates in critically ill (Chase et al., 2008a, Krinsley, 2004, Van den Berghe et al., 2001), paediatric, and non-diabetic patients (Krinsley, 2006, Van den Berghe et al., 2006a). It was associated with lower incidence of multiple organ failure (Van den Berghe et al., 2001), protection of renal function (Schetz et al., 2008, Van den Berghe et al., 2006b, Van den Berghe et al., 2006a) and reduced renal failure (Krinsley, 2004, Van den Berghe et al., 2001), reduced critical illness polyneuropathy (Van den Berghe et al., 2001), lowered incidence of blood transfusions (Krinsley, 2004, Van den Berghe et al., 2001), and reduced length of mechanical ventilation (Van den Berghe et al., 2006b) and length of stay (Krinsley, 2004, Van den Berghe et al., 2006b). It thus reduced the overall cost associated with treating the patient (Krinsley and Jones, 2006, Van den Berghe et al., 2001).

Other studies have shown benefits, such as protection of the thin endothelial cell layer lining the circulatory system (Ellger et al., 2006, Langouche et al., 2005), which in turn contributes to the prevention of organ failure (Langouche et al., 2005). Insulin therapy has been shown to suppress the inflammatory response (Das, 2003, Hansen et al., 2003) and thus be of use in reducing the incidence of infection in diabetic (Furnary et al., 1999, Pomposelli et al., 1998, Zerr et al., 1997), surgical (Van den Berghe et al., 2001), and intensive care (Van den Berghe et al., 2001) patients. It has also been shown to be of use in treating sepsis/septic shock (Das, 2003, Oddo et al., 2004), and the Surviving Sepsis Campaign recommends the use of insulin in managing septic patients (Dellinger et al., 2004). In addition, intensive insulin therapy has been shown to reduce mortality in septic patients (Ellger et al., 2008, Hansen et al., 2003), reduce renal failure (Hansen et al., 2003), and to reduce the duration of antibiotics and hyper- or hypo- thermia (Hansen et al., 2003). It is suggested that the benefits of insulin use are

associated with the reduction in hyperglycaemia, and not associated with the administration of insulin itself (Orford, 2006).

However, the reduction in mortality has not been seen in other studies (Brunkhorst et al., 2008, Finfer et al., 2009, Hirasawa et al., 2009, Liu and Mauvais-Jarvis, 2010, Morimoto et al., 2001, Preiser et al., 2009), and much higher incidence of hypoglycaemia has cast doubt on the use of insulin in critical care cohorts (Brunkhorst et al., 2008, Orford, 2006, Van den Berghe et al., 2006a). Two recent meta-analyses looking primarily at mortality (Figure 2.1) and hypoglycaemic outcomes have questioned the benefits of insulin therapy (Griesdale et al., 2009, Wiener et al., 2008), particularly in the light of a 5-6 fold increase in the risk of hypoglycaemic events (Griesdale et al., 2009, Wiener et al., 2008), which are associated with a higher mortality risk (Wiener et al., 2008).

Significant heterogeneity between studies in terms of threshold, intervention protocols, and results was noted, and thus the possibility that some patients may benefit from insulin therapy was not excluded (Griesdale et al., 2009). A significant inability of many studies to consistently meet glycaemic targets was also noted (Penning et al., 2014, Wiener et al., 2008), where consistent control to target has been associated with reduced mortality and organ failure (Chase et al., 2010b, Chase et al., 2008a) and increased odds of survival (Penning et al., 2015, Pielmeier et al., 2010b), regardless of how that control was obtained. In only one of the analyses were other outcomes (septicaemic risk and need for dialysis) considered, finding a reduction in septicaemia in ICU patients was associated with insulin therapy (Wiener et al., 2008).

2.1.2 Neonatal Intensive Care

The use of insulin therapy to treat hyperglycaemia and its effect on outcomes is not well defined in the premature NICU cohort. While short term weight gain and increased glucose tolerance have been well reported (Binder et al., 1989, Collins et al., 1991, Kanarek et al., 1991, Meetze et al., 1998, Ostertag et al., 1986, Pollak et al., 1978, Thabet et al., 2003, Vaucher et al., 1982), as well as a reduction of the incidence of sepsis (Collins et al., 1991), few studies have examined long term outcomes in this cohort. One study has shown increased 28 day mortality despite reduced hyperglycaemia, although this was not the case at the expected date of delivery (Beardsall et al., 2008). A recent study has questioned the advantage of increased weight gain with insulin therapy, proposing that it results in increased adipose tissue, and does not necessarily translate to an increase in linear growth and muscle

development (Alsweiler et al., 2012). In addition, many studies have seen a significant increase in hypoglycaemia (Alsweiler et al., 2012, Beardsall et al., 2008).

Thus, further studies are required to evaluate the potential benefits of insulin therapy to treat hyperglycaemia in NICU cohorts. In particular, it must avoid increasing the incidence of hypoglycaemia, which may confound results, and led to the early stoppage of one major trial (Beardsall et al., 2007a). Study protocols must be effective and consistent enough to determine if quality of control can be linked to clinically beneficial outcomes.

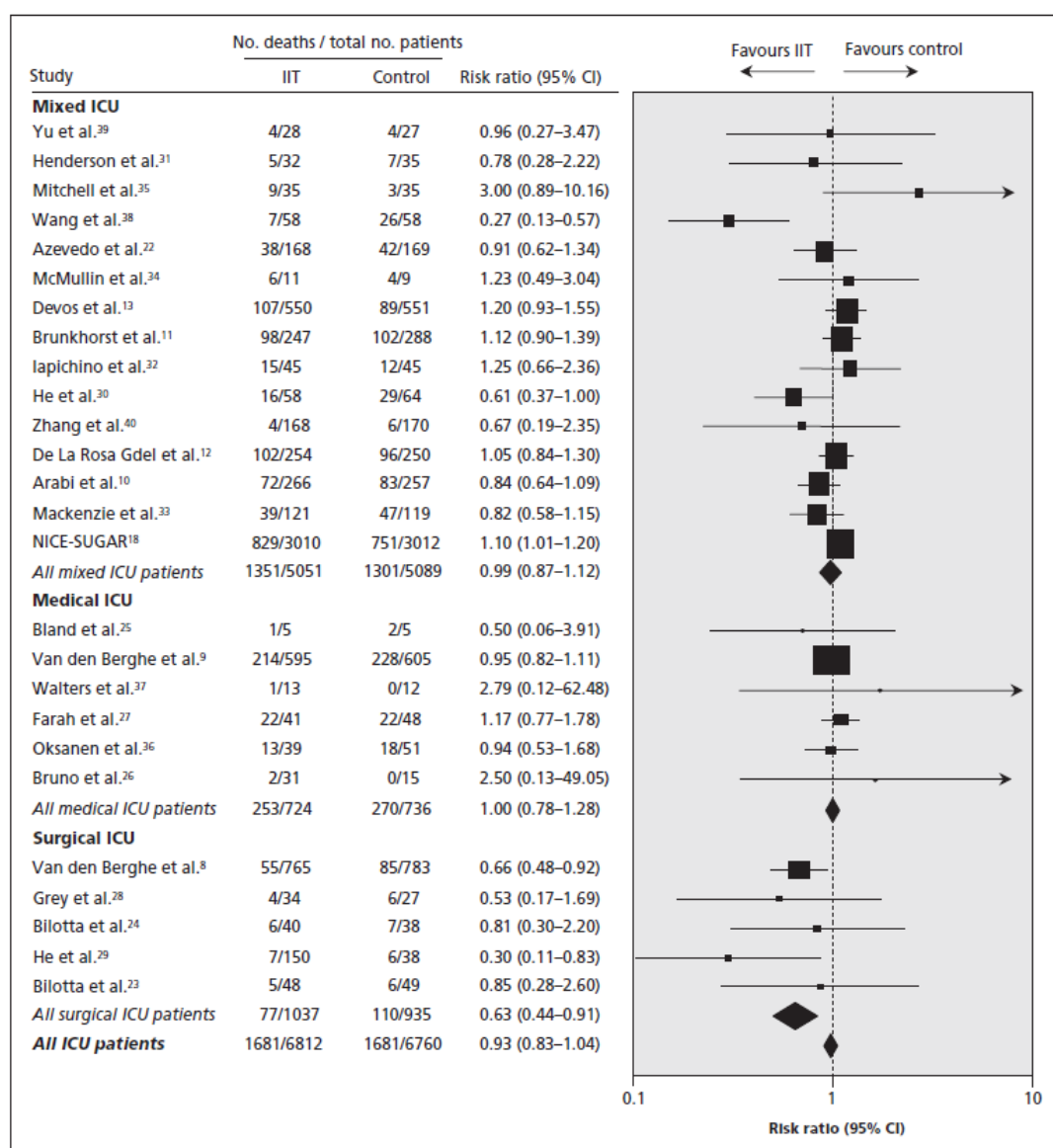


Figure 2.1: Meta-analysis of risk ratios for mortality in trials comparing intensive insulin therapy (IIT) to conventional glycaemic control. Source: Griesdale et al. (2009).

2.2 Glycaemic Control in Neonatal Intensive Care

2.2.1 Current Clinical Practice

There is also currently no global best practice method or target for glycaemic control in pre-term infants. A recent survey of neonatal hyperglycaemia in Australasia found that all of the 23 (of 27) surveyed units that responded use insulin (Alsweiler et al., 2007). Of these 23 units, 8 only use it very occasionally, 22 had a guideline or protocol for insulin use, and 17 would use glucose restriction as a first response to hyperglycaemia. Hyperglycaemia was generally defined as BG > 10 mmol/L, there was large variation in both the glycaemic control range targeted, and the glycaemic threshold for starting insulin, as shown in Figure 1.3.

In a survey of neonatal units in the Los Angeles County (Simeon and Gottesman, 1991), 80% (10 neonatal units) of those surveyed used insulin therapy to treat infants. Of these, only 2 had protocols and procedures for insulin use. In addition, 70% gave insulin to control hyperglycaemia and/or to lower serum potassium. Regarding hyperglycaemic threshold to commence insulin therapy, 60% of units that gave insulin required BG > 9.7 – 13.9 mmol/L (175 - 250 mg/dL) and/or glycosuria (elevated glucose levels in urine) before starting insulin. Methods of administration also varied between and within units, with 80% responding that they would give insulin as an IV infusions, 20% giving it as an IV bolus, 10% giving it subcutaneously, and 20% giving it intramuscular. It should be noted that some of the responding units gave insulin in more than one way. Only 50% of the units flushed the apparatus with a primer solution of insulin before administering insulin to the patient (Simeon and Gottesman, 1991), which can significantly increase the insulin delivered.

In particular, one important source of clinical error can result from insulin binding to IV tubing (Fuloria et al., 1998, Hewson et al., 2000). The percentage loss of insulin in this manner changes over time and is dependent on the flow rate, but can be as high as 90% in the first hour without priming, and 80% in the first hour if the tubing is primed first. At 8 hours, the percentage loss can be around 50% in un-primed tubing, whereas it is close to 0% when the tubing has been flushed with insulin prior to use (Fuloria et al., 1998). It is estimated that even with insulin priming of the IV tubing, consistent insulin delivery is not achieved until 6-8 hours after delivery began (Fuloria et al., 1998, Hewson et al., 2000). Thus, insulin priming and binding can have an important effect on the quality of glycaemic control by increasing certainty in the amount of insulin used.

Many studies tend to report very few statistics with respect to the quality of glycaemic control. Rates of hypoglycaemia, when discussed, are often reported as the percentage of all BG measures taken, and are typically less than 1% BG<2.2 mmol/L or 2.6 mmol/L. In addition, target ranges and definitions of hyper- and hypo- glycaemia vary between the studies. Such controversies and contradictions in literature highlight the need for an effective tool for glycaemic control in order to effectively evaluate the effect of insulin therapy on clinical outcomes in the NICU.

Lack of consensus in the definition of hyperglycaemia, target basal BG ranges, and thresholds for the initiation of insulin therapy reflect differing clinical guidelines around the practice of insulin therapy in neonatal intensive care units. Fixed or ad hoc protocols are often utilised, which lack patient specificity or rely extensively on clinical judgment. As such, protocols often fail to effectively account for variability in patient metabolic intake and response over time and between patients, resulting in poor control and oscillations between hyper- and hypo- glycaemia. This issue is confounded by errors and inconsistencies in clinical practice with respect to the physical delivery of insulin.

2.2.2 Insulin Therapy vs. Glucose Restriction

Early approaches to neonatal hyperglycaemia used caloric restriction. Nutrition regimes in the NICU primarily aim to achieve weight gain comparable to foetal growth without causing additional metabolic stress (Ditzenberger et al., 1999). Thus, the benefits of insulin therapy to promote increased glucose tolerance and weight gain have been well reported (Binder et al., 1989, Collins et al., 1991, Kanarek et al., 1991, Meetze et al., 1998, Ostertag et al., 1986, Pollak et al., 1978, Thabet et al., 2003, Vaucher et al., 1982).

While permissive underfeeding could be beneficial in the adult ICU (Arabi et al., 2011). It is not the case for neonatal intensive care, where limited storage of fuel substrates mean that glucose infusion is required to avoid hypoglycaemia (van Goudoever et al., 1993), as well as for the continued growth and development of the neonate outside of the womb (Clark et al., 2003a, Hay, 2006). The use of insulin to treat hyperglycaemia, and thus improve glucose uptake and tolerance, results in increased carbohydrate intake, and increased weight gain.

However, a recent study has questioned the advantage of this increased weight gain, proposing that it results in increased adipose tissue, and does not necessarily translate to an increase in linear growth and muscle development (Alsweiler et al., 2012). In addition, the

high incidence of hypoglycaemia observed in the tight glycaemic control group with received intensive insulin therapy led to a conclusion that the risks of insulin therapy outweigh any perceived benefits (Alsweiler et al., 2012).

2.3 Studies on Insulin Therapy and Glycaemic Control in Neonatal Intensive Care

Several studies have looked at various aspects of the use of insulin therapy to treat hyperglycaemia (Alsweiler et al., 2012, Beardsall et al., 2008, Binder et al., 1989, Collins et al., 1991, Kanarek et al., 1991, Meetze et al., 1998, Ng et al., 2005, Ostertag et al., 1986, Pollak et al., 1978, Thabet et al., 2003, Vaucher et al., 1982). While most of these have focused or based their primary result around insulin therapy versus standard treatment or glucose restriction for glucose tolerance and/or weight gain (Binder et al., 1989, Collins et al., 1991, Kanarek et al., 1991, Meetze et al., 1998, Ostertag et al., 1986, Pollak et al., 1978, Thabet et al., 2003, Vaucher et al., 1982), the following studies more specifically examined glycaemic control performance and its effect on a wider array of outcomes.

Ng et al, 2005

Ng and colleagues (Ng et al., 2005) carried out a study comparing glycaemic control in hyperglycaemic ELBW and LBW infants. Hyperglycaemia was defined as two BG > 12 mmol/L over the course of at least 4 hours. Insulin was titrated to maintain BG levels in a targeted 4-8 mmol/L glycaemic band. BG was measured hourly upon initiation of insulin therapy until BG was stable within the target band, it was measured 2-4 hourly thereafter. Treatment was stopped when the BG remained in the target band for more than 24 hours. ELBW infants required much longer periods of insulin infusion until treatment was stopped, and required more insulin to maintain euglycaemia. Hypoglycaemic incidence (BG < 2.6 mmol/L) was 15% in the ELBW and 11% in the LBW groups, which are still a relatively high incidence, although better than other studies.

NIRTURE – Beardsall et al, 2007

The NIRTURE (Neonatal Insulin Replacement Therapy in Europe) trial was a multicentre randomised trial that compared VLBW infants receiving early insulin therapy with a control group who received standard care (Beardsall et al., 2007b, Beardsall et al., 2008). The early insulin group targeted BG to 4-8 mmol/L. Infants received a fixed insulin infusion of 0.05

U/kg/hr, alongside a 20% dextrose infusion to maintain euglycaemia. Additional dextrose was given if BG dropped below 4.0 mmol/L, and insulin was discontinued if BG continued to drop. Persistent hyperglycaemia (BG>10.0 mmol/L) was treated with additional insulin or reduced dextrose. In the control group, hyperglycaemia was treated with insulin or glucose restriction if there were 2 BG>10 mmol/L. This study was stopped early because of increased incidence of parenchymal lesions in the early insulin group, and concerns regarding mortality outcomes in the study (Beardsall et al., 2007b).

Results show a high rate of hypoglycaemia (defined as BG<2.6 mmol/L for more than 60 minutes when measured by continuous glucose monitor (CGM)), with 29% of infants receiving early insulin therapy experiencing one or more hypoglycaemic events compared with 17% in the control group. There was no statistically significant difference between the early insulin and control groups with respect to morbidity and mortality rates at the expected due date. However, 28 day mortality rates were higher in the early insulin group. They concluded that there is little clinical benefit to early insulin therapy (Beardsall et al., 2007b). However, again, safety from hypoglycaemia was not obtained.

HINT – Alsweiler et al, 2014

In a randomised control trial (Hyperglycaemia and Insulin in Neonates Trial (HINT)) of tight glycaemic control (TGC) and standard care, Alsweiler and colleagues questioned whether the benefits of TGC are outweighed by the risks of hypoglycaemia (Alsweiler et al., 2012). Tight glycaemic control targeted the 4-6 mmol/L range, and the control group was only treated with insulin if the infant had BG>10mmol/L with glycosuria, and the infant was tolerating less than <100kcal/kg/day in nutritional input. Their results show greater weight gain in infants treated with insulin, with similar nutritional intake between the two groups. However, this increased weight gain did not translate into an increase in linear growth, suggesting that weight gain was due to an increase in fat mass rather than muscle or bone growth.

A doubling of the number of incidences of hypoglycaemia, where 58% of infants had one or more BG < 2.6 mmol/L in the TGC group compared with 27% in the control group, was reported, despite the fact that around a third of the control group ended up being treated with insulin as well (Alsweiler et al., 2012). Overall, the authors suggest that the risks of insulin

infusion may exceed any perceived benefits and that further trials are required to determine the relative risks and benefits.

2.4 The Problem of Patient Variability

What current studies examining insulin therapy in neonatal intensive care have failed to do is first establish an effective tool or protocol for glycaemic control. Effective, safe control needs to be achieved before examining outcomes. None of the studies discussed so far detail their glycaemic control protocol. Instead, they often refer to generic ‘titration of insulin with BG level.’

Titration of insulin with BG, or sliding scale based methods, are inherently flawed in their inability to consistently dose insulin between patients and over time. Strict sliding scale type protocols do not differentiate between patients or take into consideration differing patient condition, instead treating all patients the same all the time. In addition, they typically do not account for the nutritional intake, neglecting this important contributor to glycaemia. On the other hand, protocols that allow room for clinical judgement are biased by the attending clinician’s past and present clinical experience and context. All these prior protocols left managing variability of patient response to ad hoc clinical inputs, or simply ignored them with a one size fits all approach. What is required are protocols that explicitly account for nutrition, variability, and patient-specific response.

The advantage of model-based control is that estimations of current glucose-insulin metabolism can be made based on the measurable metabolic response to insulin. In the metabolic model presented in this thesis, the parameter insulin sensitivity (SI) describes patient specific whole-body SI, which is allowed to change over time as a patient-specific response to therapy evolves. Hence, the variability in response between patients and over time can be seen, calculated, and accounted for.

In an analysis of the effect of patient variability on glycaemic control, Le Compte et al. (2013) contrasted model based control and its treatment of SI with some of the assumptions underlying conventional protocol approaches. In contrast to patient specific and time-varying SI, fixed sliding scale protocols implicitly assume SI is constant across an entire patient cohort, as this method treats all patients the same. Similar methods, which adapt sliding scale based protocols to patient metrics such as weight (e.g. Beardsall et al. (2007a)), effectively

assume SI is constant across a patient, and does not change with time. Figure 2.2, from Le Compte et al. (2013), compares model based adaptive control to conventional methods. It is clear that for a large percentage of the time, whole cohort and per-patient constant methods fail to describe a specific patient's metabolic state. Le Compte et al. concluded that effective glycaemic control required the ability to effectively address inter- and intra- patient variability (Le Compte et al., 2013), which matches results in the adult ICU domain (Chase et al., 2011b).

It could be argued that sliding scale based protocols to some degree implicitly allow for some degree of change in SI over time indirectly through the titration of insulin to different BG levels. However, these methods are still deficient, because if this is the case they still assume the same SI within each BG range across an entire range of patients. This assumption is not accurate in the adult ICU or the NICU, as shown in (Chase et al., 2011b), and requires SI to change with BG when it is in fact a measure of response to insulin and nutrition inputs, not BG level.

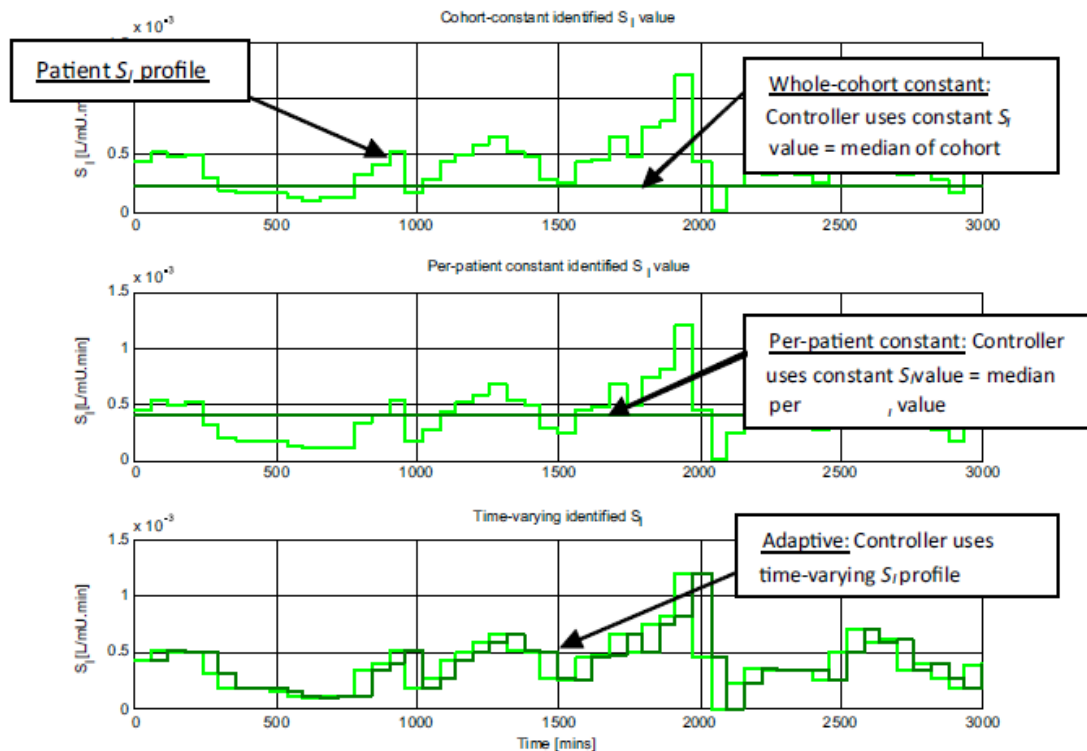


Figure 2.2: Comparison of different glycaemic control approaches and their ability to capture patient variability. The whole-cohort constant insulin sensitivity (SI) is comparable to fixed sliding scale methods, the per-patient constant SI describes sliding scale protocols that are modified by patient characteristics such as weight, and the adaptive SI describes the model based approach. Source: Le Compte et al. (2013).

In particular, Figure 2.3 was derived as part of this work from per-patient time-varying insulin sensitivity profiles over a large cohort of very/extremely premature infants in neonatal intensive care. It clearly shows that BG alone is insufficient as a marker for metabolic state, and insulin sensitivity in particular, since it is a function of insulin input, nutrition, and other interactions in patient-specific response or metabolic reactions. Therefore, effective glycaemic control must be able to make some estimate of a patient's specific metabolic state.

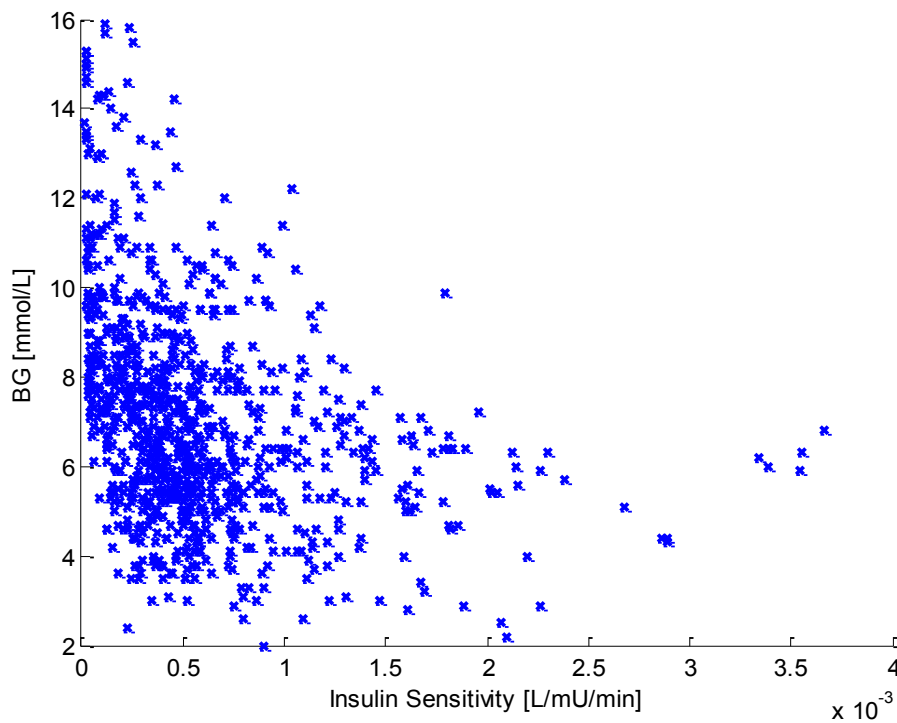


Figure 2.3: Model based insulin sensitivity (SI) is independent of blood glucose (BG) concentration.

2.5 Capturing Patient Variability Using Model Based Methods

2.5.1 Glycaemic Control Challenges in Neonatal Intensive Care

In any clinical context, for a computer model to be useful it must not require more clinical measurements than are feasible and/or cost effective. In the case of the premature infant, this requirement is of even greater importance due to their inherent fragility and very low blood volume, which limits sampling. This restriction places larger burden on a model to manage any variability between samples as patient condition evolves continuously.

More specifically, the premature infant's inherent fragility imposes many constraints on what can be measured and observed in this cohort in both clinical and research contexts. Such constraints (Farrag and Cowett, 2000) include:

- Minimal or non-invasive procedures
- Small and infrequent blood sampling due to limited blood volume
- Extrapolation of sampled data beyond the immediate context
- Study design to maximise information obtained
- Difficulty in recruiting large numbers of patients, particularly in studies with more than one trial arm.

Thus, metabolic models in neonatal intensive care must be identifiable, as well as safe and effective, using currently available clinical measurements and patient characteristics. Metabolic models must capture, as accurately as possible, the physiology relevant to the patient cohort and condition, but must also be flexible enough to be used over the range of patient specific conditions seen clinically.

2.5.2 Sources of Metabolic Variability

Glucose-insulin metabolism in infants changes over time, and between patients. In particular, it may be different from adult metabolism, justifying independent models for glycaemic control. These models can be based upon, or adapted from, adult models. However, they will have clear cohort specific differences, although the underlying fundamental metabolic pathways and dynamics are the same.

In addition, to best capture variability over time and differences between patients, the model should take into account a number of possible sources of metabolic change. This change may arise from: endogenous glucose production, central nervous system uptake, endogenous insulin secretion, insulin clearance, and glucose absorption in the gut. All of these differences can result from evolving patient specific condition and stress response.

In particular, differences in glucose-insulin metabolism between patients can potentially be captured through patient descriptors, such as weight, age, and condition. Changes in glucose-metabolism over time may also be described as a function of patient condition, nutritional and medicinal input, and blood species. Thus, the more accurate and effective the model, the more model-based SI changes reflect actual changes in peripheral and whole body SI. If SI

changes can be tightened to predominantly reflect changes, then control can be tighter and safer.

Endogenous insulin secretion

From early in the gestational period, the foetal pancreas is a major source of insulin (Adam et al., 1969b). However, bi-phasic insulin secretion in response to nutrient release may not begin to mature until after birth (Otonkoski et al., 1988). For this reason, it is reasonable to assume that insulin secretion in premature infants is different from the adult case, and justifies its own model. However, like the adult case, there are no such models in the literature.

Insulin secretion in preterm babies has only been indirectly analysed by observing plasma insulin concentrations. However, insulin is cleared by both the kidneys and the liver in a highly variable manner, as well as being cleared in peripheral tissues. These factors ensure that plasma insulin concentrations are an inaccurate measure of insulin secretion. Pancreatic insulin secretion has been estimated in adults using models of C-peptide kinetics (Eaton et al., 1980, Polonsky et al., 1986, Van Cauter et al., 1992), as C-peptide is secreted in equimolar quantities with insulin, but is only cleared by the kidney. Therefore, the much simpler kinetics of C-peptide provide a more accurate method of estimating insulin secretion rates (Van Cauter et al., 1992), but have not been used in the past with neonates.

Insulin clearance pathways

Insulin is cleared via a number of pathways, including the liver, kidneys, and peripheral degradation. Glomerular filtration rate (GFR) is commonly used to assess renal function. GFR increases with gestational and postnatal age in premature infants (Coulthard, 1985), and rises during childhood through to puberty (Lebenthal and Lebenthal, 1999). It is likely that these changes in GFR translate to changes in the kidney clearance of insulin with age. In addition, several studies have also shown that plasma insulin levels are higher in preterm than term infants (Broussard, 1995, Pollak et al., 1978, Tyralla et al., 1994), reflecting a whole body difference in clearance dynamics. Thus, prematurity specific models of insulin clearance would enable more accurate modelling of insulin concentrations, and thus better assessment of SI for safer dosing.

Endogenous glucose

Endogenous glucose production (EGP) plays a central role in glucose homeostasis in the body. Glucose production occurs mainly in the liver, and its main processes are gluconeogenesis and glycogenolysis. These processes are affected by prematurity. In particular EGP is observed to persist in premature infants (Cowett et al., 1983, Farrag et al., 1997), which contrasts the normal adult case (Wolfe et al., 1979). EGP in very premature infants has been shown to be responsive to exogenous glucagon (Van Kempen et al., 2003b), but not exogenous administration of amino acids (Diderholm et al., 1999, Van Kempen et al., 2003a) or plasma insulin (Suneag et al., 1994b). It can also to vary with patient weight and/or gestational age (Keshen et al., 1997, Van Kempen et al., 2003b). There is thus scope and potential to build EGP models to describe the unique physiology of the premature infant and capture sources of inter- and intra- patient variability.

Insulin-independent central nervous system uptake

In the adult, the brain accounts for approximately 50% of the bodies glucose uptake (Triplitt, 2012), so it must be accounted for in any model-based approach. In a metabolic model of adult critical care insulin-glucose dynamics (Lin et al., 2011) central nervous system (CNS) uptake is modelled as population constant, which reflects the relatively constant brain mass across individuals and resulting constant glucose metabolism by the brain (Gruetter et al., 1998). However, in infants CNS increases with increasing gestational age (Koh et al., 1988b), and infants have larger brain-body weight ratios than adults (Farrag et al., 1997, Hertz et al., 1993). So brain weight, and thus CNS uptake, may be better predicted by patient-specific head circumference (Cornblath et al., 1990).

Glucose absorption in the gut

The small intestine is a major site of nutrient and water absorption. During the last trimester of pregnancy the foetal intestine lengthens (Aynsley-Green, 1983), and enzyme activity increases (Lebenthal and Lebenthal, 1999). However, mature levels of digestive enzyme secretion are not reached until the end of foetal gestation (Rouwet et al., 2002), and lactase secretion is only ~30% of mature secretion at 34 weeks (Lebenthal and Lebenthal, 1999). Gastrointestinal motility is also not mature until the 40th week gestational age (Berseth, 1996). Further, exposure of the neonatal small intestine to enteral feeds results in changes in mucosal structure and function (Aynsley-Green, 1983). Thus gastro-intestinal permeability

changes with gestational and post natal age (Rouwet et al., 2002), with absorption capacity most limited in neonates born <28 weeks gestational age (Rouwet et al., 2002), and is thus a further source of variability.

2.6 Summary

Insulin therapy is widely used to treat hyperglycaemia in intensive care. While early studies in the adult ICU showed significant benefits and reduced mortality associated with the use of insulin therapy, some later studies have not been able to replicate these results, and controversy remains. In most cases, both in adults and neonates, insulin therapy is associated with significantly higher incidence of hypoglycaemia, which carries significant attendant risks.

In neonatal intensive care, the literature surrounding the effect of insulin therapy on outcomes is relatively scarce, and reflects similar mixed results and controversy. However, what current studies fail to do is to first develop a tool for safe and effective glycaemic control, before trying to establish the effect on outcomes. Hence, there is a need for better dosing methods before being able to assess the impact of glycaemic control on clinical outcomes.

There is no universal or widely accepted protocol for insulin dosing, nor threshold for starting intervention. In addition, clinical titration methods in all reported studies fail to adequately account for variability between patients and within patients over time. Model based approaches offer this capability.

Potential sources of metabolic variability that such models need to capture include endogenous glucose production, central nervous system uptake of glucose, pancreatic insulin secretion, insulin clearances in the body, and gut absorption of glucose. This thesis aims to analyse and create the first models of these aspects of glucose-insulin metabolism in the preterm neonate to better capture inter- and intra- patient variability in response to insulin, and thus enable safer, more effective glycaemic control.

2.7 Preface

The remaining thesis chapters cover this topic as follows:

Chapter 3 presents common metabolic models of the glucose insulin system, and introduces the basic form of the model developed throughout this thesis.

Chapters 4 and 5 examine insulin secretion and clearance in premature neonates using unique clinical data from a VLBW cohort. Gender and BG based models of insulin secretion are developed, and insulin clearance pathways are identified and modelled.

Chapter 6 explores the contribution of endogenous glucose production to glycaemic control, and develops models based on literature data and glycaemic control outcomes in simulation.

Chapter 7 examines non-insulin mediated pathways for glucose clearance, focusing in particular on the central nervous system uptake and estimation of this based on brain mass and head circumference.

Chapter 8 uses continuous glucose monitor (CGM) data in premature and term infants to estimate glucose absorption in the gut.

Chapter 9 summarises the model developed in Chapters 4 to 8.

Chapter 10 uses clinical data and stochastic modelling to develop and optimise a model-based protocol for glycaemic control in the NICU.

Chapter 11 presents clinical results from the protocol developed in Chapter 10.

Chapter 12 analyses SI and its variability based on clinical cohorts, birth weight, and gestational age.

Chapter 13 and 14 summarise the conclusions drawn from this thesis, and examine potential avenues for future work.

Chapter 3: Model structure

Physiological models of the glucose-insulin system have been developed with differing levels of complexity for a wide range of scientific and clinical applications. Some are utilised for the determination of model-based measures of physiology, such as insulin sensitivity, or to accurately capture or determine aspects of human metabolism and physiology. Such models tend to be more complex and comprehensive, requiring higher data density and/or measurements of multiple metabolic species. Other models are designed for specific clinical applications, such as glycaemic control in intensive care, or Type 1 Diabetes cohorts. Such models tend to be less complex, as metabolic measurements are minimised in clinical settings due to cost, availability, time constraints, clinical workload, or patient condition and comfort. Thus, model capabilities are determined not only by the degree to which they accurately reflect physiological processes, but also the context within which they are designed and implemented (Chase et al., 2011a).

One very common form of physiological model is the compartment model. The compartment model is physiologically intuitive, as it consists of a series of 'compartments' where model species exist at some concentration. Changes in compartment concentrations are brought about by the appearance and disappearance of species from these compartments. In a physiological sense, species can appear via secretion or exogenous administration, and appear and/or disappear due to diffusion/transport between compartments or interactions between substances. Species can disappear via known clearance mechanisms, such as glomerular filtration in the kidney, or via degradation in their metabolic utilisation. In this way, compartment based models can be designed to capture key dynamics to a chosen level of resolution, suitable to their intended use.

Choosing the degree of resolution of a model is important. Such models must capture key physiological dynamics to be physiologically valid and relevant within their application. However, they must also be mathematically identifiable (Docherty et al., 2011) and practically applicable given the available measurement within their chosen application (Chase et al., 2006). In clinical practice, a key constraint is the availability of clinical data in terms of both measurement type and frequency. Limited data availability limits parameter identification capabilities. The primary aims of a physiological model in critical care is to improve the quality of care without an unnecessary increase in clinical workload and cost of

care, a very difficult trade-off. This trade-off typically eliminates the ability to add measurements to enhance the model application.

While several models have been developed for use in adult critical care or diabetes, physiological modelling of neonatal glucose-insulin metabolism is far less common. To date, only Le Compte (Le Compte, 2009) has developed a model for glycaemic control in premature infants. This chapter will briefly review several glycaemic control approaches in adult critical care as a basis for general control approaches, and will present the model of Le Compte (2009) as a starting point for further model development in this thesis.

3.1 Metabolic Models in Critical Care

Several approaches to modelling and/or managing glucose-insulin metabolism in adults have been developed (Bergman et al., 1979, Chee et al., 2003, Hovorka et al., 2004, Kovatchev et al., 2009, Lehmann and Deutsch, 1998, Pielmeier et al., 2010b, Sorenson, 1985, Van Herpe et al., 2006). Typical examples of approach to glycaemic control include Chee et al. (2003), Hovorka et al. (2004), and Bergman et al. (Bergman et al., 1985, Bergman et al., 1979). These models/methods are reviewed more extensively elsewhere (Chase et al., 2006, Le Compte, 2009, Lin, 2007), but differ in structure and implementation, each with different strengths and weaknesses.

The approach developed by Chee et al. (2003) is based on traditional control theory. While the results show a significant tightening of glycaemic control, mean BG remained high (Chee et al., 2003). This control method is not based on a physiological modelling, and thus must be tuned to different cohorts, a process that is likely unintuitive.

Hovorka et al. (2004) developed a sophisticated physiological model for glycaemic control in Type 1 Diabetes, and a modified version for glycaemic control in critical care (Plank et al., 2006). This model is compartment based, with 9 population constants, and 6 patient specific constants. The relative complexity and physiological comprehensiveness of this model reflect its origins in physiological research (Hovorka et al., 2002). However, practical identifiability may be a problem in critical care settings with this model and minimal blood sampling due to workload or other clinical concerns.

The minimal model of Bergman et al. (Bergman et al., 1985, Bergman et al., 1979) has been extensively utilised and modified over time. Many compartment models used for glycaemic

control or studies of metabolic physiology have some degree of origin in it (Chase et al., 2006). It forms the basis for the ICING model (Evans et al., 2011, Lin et al., 2011) used for glycaemic control in adult intensive care, and the NICU model developed for use in neonatal intensive care (Le Compte, 2009, Le Compte et al., 2009, Le Compte et al., 2010a).

Before adoption as a standard of care, model based control should be extensively tested for safety and effectiveness. Clinically, such testing requires extensive clinical time and resources, and may pose a risk to the patient. Alternatively, extensive testing can be carried out using virtual patients, but such testing is most effective when virtual patients based on clinical data and metabolic models are used (Chase et al., 2007, Chase et al., 2010a). It is equally important that such virtual patients be validated (Chase et al., 2010a).

Overall, many such models exist, but few have been used clinically. Equally, virtual patients can be used to optimise design, but often lack validation in clinical or other use. Thus, models offer potential, but are often not as clinically feasible or relevant, as needed.

3.2 Neonatal Glycaemic Modelling

In the case of the very/extremely preterm neonate, to date, only Le Compte et al. have attempted to model the glucose-insulin regulatory system of this cohort (Le Compte, 2009, Le Compte et al., 2010a). This NICU model is structurally based on the model by Lin (2007) and is shown pictorially in Figure 3.1. Its parameters are adapted to reflect premature infant physiology, and it contains additional terms for endogenous glucose production (*EGP*), central nervous system uptake of glucose (*CNS*), and insulin secretion (structure: $I_B e^{-x}$). These models utilise a time varying whole body insulin sensitivity (*SI*) parameter to describe changes in patient metabolic condition.

The NICU model of Le Compte et al. (Le Compte, 2009, Le Compte et al., 2010a) has 3 main compartments in Figure 3.1 (*G*(*t*), *I*(*t*), and *Q*(*t*)), which are mathematically defined:

$$\begin{aligned} \dot{G} = & -p_G G(t) - S_I G(t) \frac{Q(t)}{1 + \alpha_G Q(t)} + \frac{P_{TPN}(t) + P_{EN}(t)}{V_{g,frac}(t) m_{body}} \\ & + \frac{(EGP - CNS m_{brain,frac}) m_{body}}{V_{g,frac}(t) m_{body}} \end{aligned} \quad (3.1)$$

$$\dot{I} = - \frac{n I(t)}{1 + \alpha_I I(t)} - \frac{u_{ex}(t)}{V_{I,frac} m_{body}} + I_B e^{-\left(k_I \frac{u_{ex}(t)}{V_{I,frac} m_{body}}\right)} \quad (3.2)$$

$$\dot{Q} = k(I(t) - Q(t)) \quad (3.3)$$

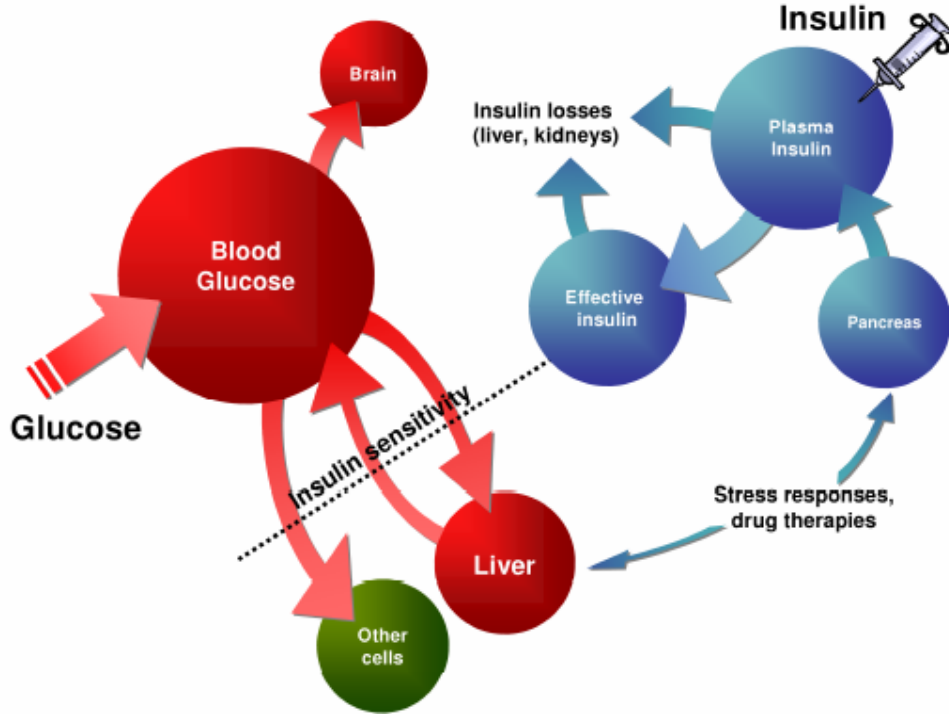


Figure 3.1: NICU model of glucose-insulin metabolism. Arrows show movement of glucose (red) and insulin (blue) into and out of central compartments. Insulin sensitivity mediates the uptake of glucose by the liver and other body cells.

In the blood glucose compartment (G), p_G is non-insulin mediated clearance of glucose (e.g. by kidneys), S_I is the insulin sensitivity that drives insulin-mediated uptake of glucose, α_G saturates the insulin-mediated uptake of glucose, P_{TPN} is the exogenous intravenous glucose appearance, P_{EN} is glucose appearing via the enteral route, EGP is the basal endogenous glucose production, CNS is the non-insulin mediated central nervous system uptake. $V_{g,frac}$ is the volume of distribution of glucose, m_{body} is the mass of the body, and $m_{brain,frac}$ is the fraction of brain to body mass.

In the plasma insulin compartment (I), $u_{ex}(t)$ is the exogenous insulin input, I_B is basal insulin secretion, which is modified by an exponential representing the suppression of insulin secretion in the presence of exogenous insulin, as determined by k_I . The rate parameters n and k determine the clearance of insulin from the system and the transport between the plasma (I) and interstitial compartments (Q) respectively. $V_{I,frac}$ is the distribution volume of insulin, and α_I saturates insulin clearance.

Absorption of glucose via the enteral route was modelled:

$$P_{EN}(t) = \bar{P}_{EN,i+1} + (P_{EN}(t_i) - \bar{P}_{EN,i+1})e^{-k_p(t-t_i)} \text{ for } t_i < t < t_{i+1} \quad (3.4)$$

where the effective half life of glucose transport from gut to plasma (k_p [min^{-1}]) is defined according to whether feed rates are increasing (k_{pr}) or decreasing (k_{pd}):

$$k_p = \begin{cases} k_{pr} & \text{where } P_{EN,i+1} > P_{EN}(t_i) \\ k_{pd} & \text{where } P_{EN,i+1} < P_{EN}(t_i) \end{cases} \quad (3.5)$$

3.3 The Intensive Care Insulin-Nutrition-Glucose (ICING) Model

Both the adult ICU model by Lin (Lin, 2007) and the NICU model by Le Compte (Le Compte, 2009) sought to balance model simplicity with physiological descriptiveness. A later iteration of the adult ICU model is the Intensive Care Insulin-Nutrition-Glucose (ICING) model (Lin et al., 2011), which was developed to include greater physiological descriptiveness, particularly around the clearance pathways of insulin, without sacrificing identifiability and clinical applicability.

The basic ICING model structure, shown pictorially in Figure 3.2, uses some of the main compartments and is defined:

$$\dot{G} = -p_G G(t) - S_I G(t) \frac{Q(t)}{1 + \alpha_G Q(t)} + \frac{P_{ex}(t) + EGP_b - CNS}{V_g} \quad (3.6)$$

$$\dot{I} = -\frac{n_I I(t)}{1 + \alpha_I I(t)} - n_K I(t) - n_I (I(t) - Q(t)) + \frac{u_{ex}(t)}{V_I} + (1 - x_L) \frac{u_{en}}{V_I} \quad (3.7)$$

$$\dot{Q} = n_I (I(t) - Q(t)) - n_c \frac{Q(t)}{1 + \alpha_G Q(t)} \quad (3.8)$$

$$\dot{P}_1 = -d_1 P_1 + P(t) \quad (3.9)$$

$$\dot{P}_2 = -\min(d_2 P_2, P_{max}) + d_1 P_1 \quad (3.10)$$

$$P_{ex}(t) = \min(d_2 P_2, P_{max}) + PN(t) \quad (3.11)$$

$$u_{en}(t) = k_1 e^{-I(t)k_2/k_3} \quad (3.12)$$

Structurally, Equation 3.6, which describes the rate of change of BG, is similar to Equation 3.1. However, the simple insulin clearance dynamics (driven by n and k) of Equation 3.7 are more physiologically comprehensive, including first part hepatic extraction of pancreatic

insulin secretion (x_L), insulin saturated clearance of insulin by the liver (n_L), kidney clearance (n_K), and diffusion of glucose between the plasma and peripheral compartments (n_I). The volumes of distribution of insulin in plasma is V_I . Pancreatic insulin secretion (u_{en}) has a base rate, k_1 , which is suppressed by parameters k_2 and k_3 .

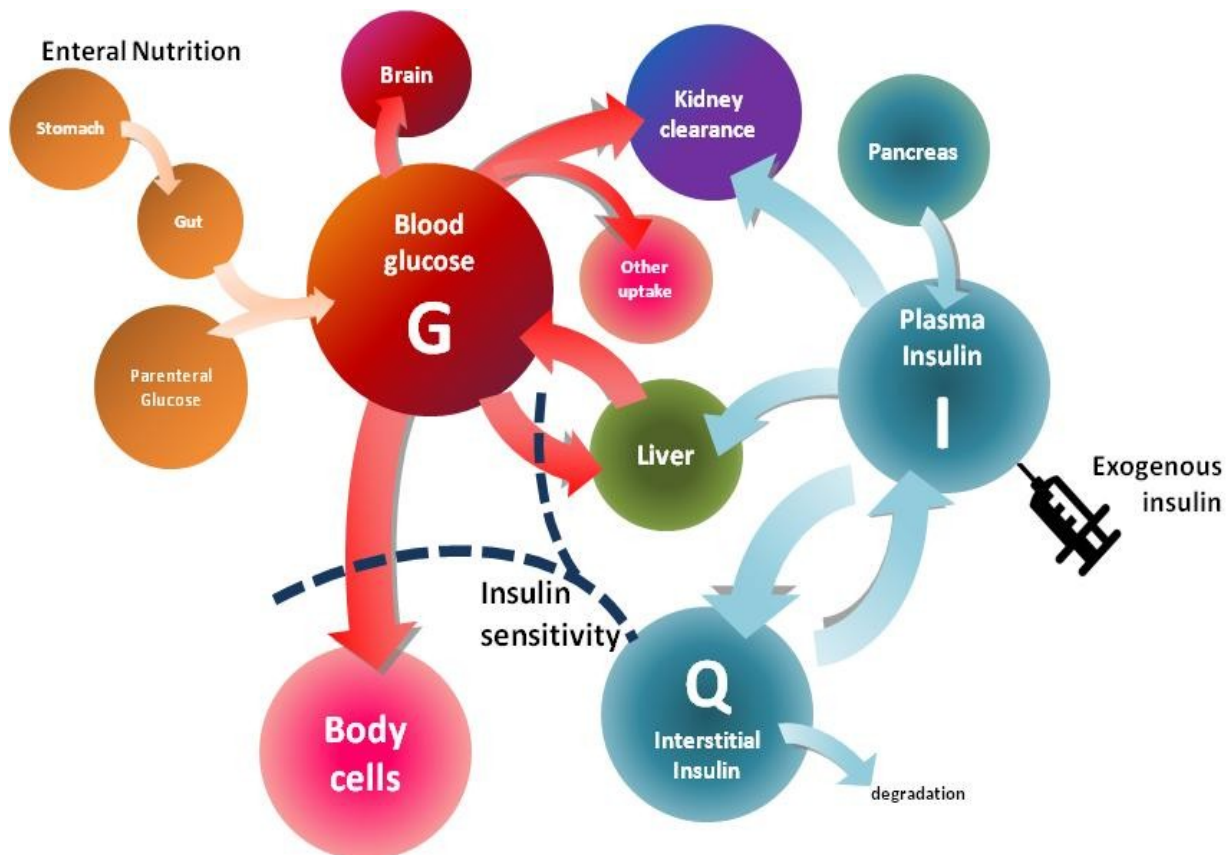


Figure 3.2: Pictorial description of the ICING model. Arrows show movement of glucose through gastro-intestinal compartments (orange) and into and outcome a central blood glucose compartment (red). Blue arrows show insulin movement between plasma and interstitial compartments, as well as secretion and clearance.

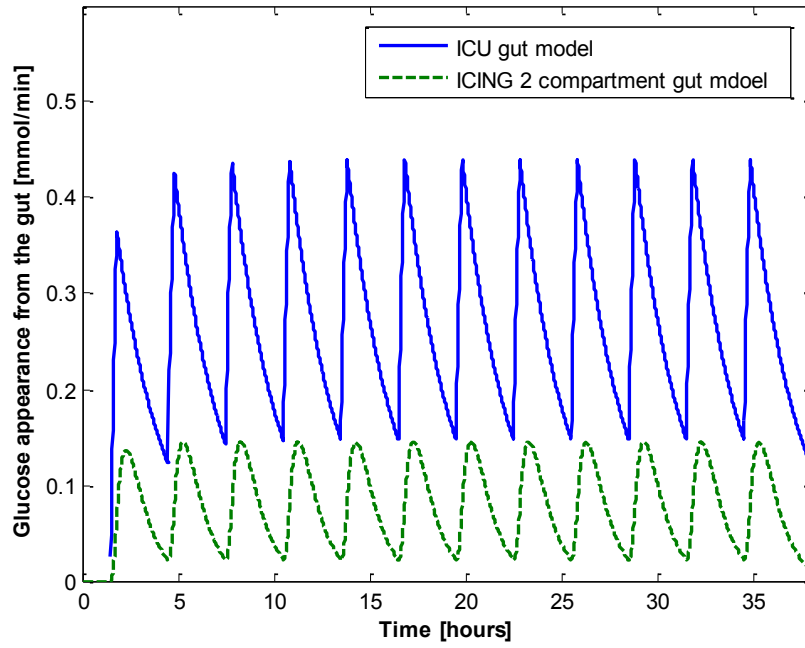


Figure 3.3: Comparison of two models for glucose appearance from enteral feeds. Figure derived from enteral feed data for a preterm infant, using adult d_1 and d_2 parameters.

Of note, is the change in the model for gastro-intestinal processing and absorption of insulin. One of the main issues with Equations 3.4 and 3.5 is that they do not necessarily conserve mass. The ICING model uses a simple 2 compartment gut model from Dalla Man et al. (2006) to describe the absolute amount of glucose ([mmol]) in the stomach (P_1) and small intestine (P_2). Glucose from an enteral feed (P) is transferred via gastric emptying from the stomach to the intestine (d_1). Absorption from the intestine to the blood stream is driven by the rate constant d_2 , with a maximal glucose transport rate of P_{max} . A comparison of the two models is shown in Figure 3.3, where the failure of the NICU gut model to conserve mass is clearly apparent.

3.4 Insulin Sensitivity and Model Based Control

The model-based glycaemic control framework used in this thesis is known as STAR (Stochastic TARgeted) because of its use of stochastic forecasting models. Stochastic models of likely future changes in SI are used to actively target likely BG outcomes to a clinically defined target range (Evans et al., 2011, Le Compte et al., 2009, Luque et al., 2009). In this model based framework, SI is patient-specific and time-varying, describing a patient's current metabolic state. This model parameter is identified on an hour-to-hour basis from

clinical data using integral-based fitting (Hann et al., 2005). In addition to being a marker of peripheral insulin sensitivity, SI also captures uncertainty around patient-specific endogenous insulin and glucose production (Chase et al., 2011b, Dickson et al., 2013).

A detailed description of stochastic modelling and validation can be found in (Lin, 2007, Lin et al., 2008). In summary, kernel density estimate methods are applied to matched pairs of SI now and SI in a defined number of (k) hours time (e.g. $\{SI_n, SI_{n+k}\}$), to generate a continuous probability distributions across the range of observed SI based on a cohort of identified SI values. Thus, for any identified SI_n the stochastic model provides a probability density function describing likely values of SI_{n+k} , on a cohort basis over the next k hours.

These distributions of likely insulin sensitivity outcomes can then be used to generate BG outcome distributions for a given insulin and nutrition treatment, as shown in Figure 3.4. Insulin treatment can then be selected based on likely BG outcomes, using defined targets and thresholds. In particular, hypoglycaemic risk can be quantified (usually as the $\%BG < 4.0$ or 4.4 mmol/L) and directly managed (Evans et al., 2011, Fisk et al., 2012), which is unique to this approach.

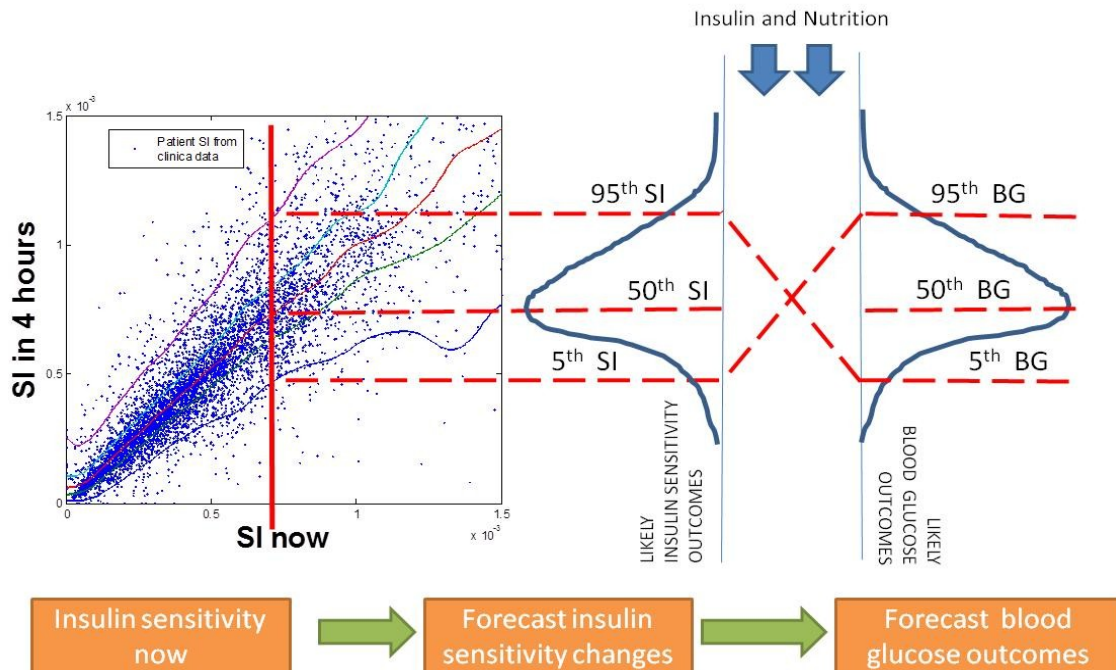


Figure 3.4: Stochastic forecasting of insulin sensitivity (SI) and blood glucose (BG) outcomes. First a patient's current SI is determined. Distributions of future insulin sensitivity are generated from stochastic models, and these are used to generate distributions of BG outcomes for a given insulin/nutrition treatment (usually characterised by the 5th, 50th, and 95th percentiles).

Stochastic models are generated on a cohort-specific basis, and the median likely change in SI is no change. However, any single patient is not guaranteed to follow the whole-cohort defined behaviour. In practice, it usually means the stochastic model is conservative for many patients, with wider 5th - 95th percentile bands than may be seen on a patient specific basis. Inevitably, this conservatism has an impact on the tightness of control. However, it also allows unusually variable patients, who are most at risk of hypoglycaemia, to be treated in a safe manner. Thus, this approach to model creation, as well as its application, favours safety over performance.

Generation of stochastic models requires sufficient data density (minimum of ~1300 patient hours) (Lin et al., 2006). Thus, sub-cohort defined stochastic models based on patient characteristics for specific groups are not always possible. Such sub-cohort models could offer greater precision and specificity to control. Stochastic models must also balance the desired tightening of percentiles, perhaps by sub-cohort definition, with the increased risk for patients who are not well defined by any sub-cohort.

3.5 Summary

Physiological models must be physiologically valid, mathematically identifiable, and clinically useful. This chapter presented the NICU model for glycaemic control in neonatal intensive care, and the ICING model for glycaemic control in adult intensive care. These models are used as a starting point from which a Neonatal Intensive Care Insulin-Nutrition-Glucose (NICING model) will be developed. Adaptation of the ICING model with respect to neonatal physiology will revisit the modelling approach of Le Compte et al. (Le Compte, 2009, Le Compte et al., 2010a), and attempt to more comprehensively model certain aspects of neonatal insulin-glucose metabolism, moving beyond simple population constants where possible, feasible, and relevant. This modelling will be based on meta-analyses of published literature data, as well as a data cohort of relatively rare and abundant clinical measurements made available from a large scale glycaemic control trial in very premature infants (Alsweiler et al., 2012).

Chapter 4: Modelling Insulin Secretion

4.1 Introduction

Adequate modelling of endogenous (pancreatic) insulin secretion plays an important part in model-based glycaemic control. During intervals when no exogenous insulin is being delivered, pancreatic insulin secretion is the only source of insulin, and thus has a large impact on identified, model-based SI. When exogenous insulin is being delivered, pancreatic insulin secretion makes up a small proportion of all insulin appearance, thus affecting this SI to a much smaller degree. Proper modelling of insulin secretion can reduce model identified SI variability, as patient-specific perturbations from modelled dynamics are reduced. As a result, identified, model-based SI would more accurately reflect peripheral SI, and overall glycaemic control could be improved.

All prior studies into neonatal insulin secretion have relied on plasma insulin concentration alone as a qualitative surrogate indicator of increased insulin secretion (e.g. Andronikou and Hanning (1987), Cser and Milner (1975), Grasso et al. (1975), Grasso et al. (1990), Grasso et al. (1973), Grasso et al. (1968), Grasso et al. (1970), Reitano et al. (1978)). Importantly, in all these studies, there has been little or no discussion of clearance kinetics or other factors impacting the results when using this approach. However, insulin is highly unreliable in this role because of its multiple clearance pathways, as well as the variability in the clearance rate of these pathways between patients and over time.

Pancreatic insulin secretion has typically been estimated in adults using models of C-peptide kinetics (Eaton et al., 1980, Polonsky et al., 1986, Van Cauter et al., 1992). C-peptide is secreted in equimolar quantities with insulin, but is only cleared by the kidney, a well known and measureable clearance pathway. C-peptide concentration is thus a more accurate predictor of insulin secretion compared with plasma insulin concentration due to its simpler, less variable, and more measureable clearance kinetics (Van Cauter et al., 1992). Thus, a very important aspect of the dataset in this chapter is the availability of C-peptide measurements, a protein that relatively few studies have reported for this population due to sampling difficulties and limits in neonates because of their very small blood volume.

This aim of this study was to determine insulin secretion in premature neonates, calculated from plasma C-Peptide concentrations. It is based on clinical data from premature babies

enrolled in a randomised controlled trial of tight glycaemic control (Alsweiler et al., 2012). A particular goal is to determine predictors for insulin secretion in hyperglycaemic preterm babies, and create a model suitable for use in glycaemic control.

4.2 Methods

4.2.1 Clinical Cohort

Plasma samples were collected from 88 preterm babies during a prospective, randomised trial of tight glycaemic control of preterm babies, the HINT trial, (Alsweiler et al., 2012) Australian Clinical Trials Registry 12606000270516, ethics approval from the New Zealand Northern X Ethics Committee. This trial compared tight glycaemic control (TGC) using insulin to maintain the BG 4-6 mmol/L in hyperglycaemic preterm babies, with standard care (control group), at the National Women's Health NICU in Auckland, New Zealand (Alsweiler et al., 2012). Written informed consent was obtained from a parent of each child.

Babies were eligible for enrolment if they were born at <30 weeks gestational age (GA), or birth weight (BW) <1500g, and had become hyperglycaemic, defined as two consecutive BG>8.5 mmol/L at least 4 hours apart. Babies were randomised when they became hyperglycaemic, which was usually 4-5 days postnatal age (PNA). Small for gestational age (SGA) was defined as less than the 10th percentile on population based growth charts. Birth weight z scores were calculated based on a similar published data cohort (Beeby et al., 1996).

Of the 88 babies recruited to the trial, 41 were of GA<32 weeks and had sufficient plasma from 1 or more of the samples for further retrospective analysis, totalling 54 samples taken at a median of 7 days after study randomisation. Of these 41 babies, 20 were from the TGC group, contributing 25 samples. Median BW and GA was 839g and 27 weeks respectively. Demographic details are given in Table 4.1.

Nutritional intake, BG, growth and insulin infusions were recorded. Parenteral nutrition was commenced within the first day after birth and enteral feeds with small amounts of breast milk were usually started in the first one to two days after birth. Lactose is comprised of both glucose and galactose, which are of approximately equal molar mass, with only glucose directly contributing to BG concentration. Therefore, glucose appearance from lactose was estimated as half the available lactose.

Table 4.1: Characteristics of babies randomised to the HINT trial whose plasma samples were analysed for plasma C-Peptide concentrations. Numbers are presented as median [IQR] or number (% of total). CRIB 2: Clinical Risk Index for Babies.

Characteristic	Value
Total patients	41
Control group	21
Tight Glycaemic Control group	20
Male (%)	20 (49%)
Multiple Birth	11 (27%)
Antenatal glucocorticoid exposure	39 (95%)
Maternal diabetes	1 (2%)
Gestational age, weeks	27.2 [26.2 - 28.7]
Birth weight (grams)	839 [735 – 1000]
Birth weight Z score	-0.19[-1.03 - 0.14]
Small for gestational age	6 (15%)
CRIB 2 score	12 [10-14]
Ethnicity	
Asian	9 (22%)
Caucasian	11 (27%)
Maori	17 (41%)
Pacific Island	4 (10%)

Blood samples were taken at randomisation (DOR=0), 7 and 14 days post-randomisation (DOR=7 and DOR=14 respectively), and at 36 weeks GA. Plasma insulin, insulin-like growth factor 1 (IGF I), and glucose concentrations were measured, and remaining plasma samples were frozen. BG measures were taken when clinically indicated and determined using a glucose oxidase method (ABL 700, Radiometer Ltd, Copenhagen, Denmark). Plasma insulin concentrations were measured on an Azsym system auto-analyzer (Abbott Laboratories, Abbott Park, IL) (Alsweiler et al., 2012). Retrospective C-peptide analysis was carried out on samples with sufficient remaining plasma from samples taken 0-15 days after randomisation at a GA <32 weeks. Plasma C-peptide concentrations were determined using immunometric assays (Elecsys 2010, Roche Diagnostics, Germany).

4.2.2 C-peptide Model

Plasma C-peptide concentrations were used to estimate insulin secretion, using a well-accepted 2 compartment kinetics model: (Van Cauter et al., 1992)

$$\frac{dC}{dt} = S - (k_1 + k_3)C + k_2Y \quad (4.1)$$

$$\frac{dY}{dt} = k_1 C - k_2 Y \quad (4.2)$$

where C is the amount of C-peptide in the central compartment comprising of plasma and tissues in rapid equilibrium with the plasma, and Y is the amount of C-peptide in the peripheral extra vascular compartment. C-peptide and insulin are secreted in equimolar amounts into the central compartment at rate S . Transport of C-peptide from the central to the peripheral compartment, and back, is described by k_1 and k_2 . Irreversible renal clearance of C-peptide from the central compartment via the kidney is modelled by k_3 (Van Cauter et al., 1992).

Sampling constraints due to limited blood volume in this cohort mean frequent, serial measurements of C-peptide, as used in adult studies, were not clinically or ethically possible. A steady-state assumption can be made to enable deconvolution of the insulin secretion rate from measured C-peptide concentrations. Under this steady-state assumption, it follows from Equation 4.2 that the rate of C-peptide entering and leaving the peripheral compartment must be equal. Hence, substituting this equality into Equation 4.1 and rearranging yields:

$$S = k_3 C \quad (4.3)$$

Equation 4.3 holds whether C is an absolute amount [pmol] or concentration [pmol/L]. Since insulin is secreted in equimolar quantities with C-peptide, under steady state conditions the rate of secretion of insulin is directly proportional to the measured concentration of C-peptide in the central compartment.

4.2.3 C-peptide Kinetic Parameters in Preterm Neonates

Since no studies have been performed in preterm or term neonates to determine C-peptide kinetics, adult data and methodology (Polonsky et al., 1986, Van Cauter et al., 1992) were used as an approximation, given that the fundamental physiological kinetics and functionality are also no different. The reported values and ranges for each of the kinetic parameters of Equations 4.1 and 4.2 are given in Table 4.2.

A short half life of 4.95 min and a fraction, F , of 0.76 was used based on non-obese and non-diabetic adult data. The long half life thus was calculated using (Van Cauter et al., 1992):

$$long - half\ life\ (min) = 0.14 * age\ (years) + 29.2. \quad (4.4)$$

Table 4.2: Kinetic C-peptide parameter ranges in adults. Data sources: (Bier, 1998, Eaton et al., 1980, Van Cauter et al., 1992). All values are mean \pm SEM

	Patient Cohort	Short half life [min]	Long half life [min]	Fraction F	k_1 [min ⁻¹]	k_2 [min ⁻¹]	k_3 [min ⁻¹]
Eaton et al, 1980	Normal n=20	-	-	-	0.047 \pm 0.002	0.035 \pm 0.002	0.049 \pm 0.001
Van Cauter et al, 1992	Normal n=111	4.95 \pm 1.04	32.4 \pm 5.0	0.76 \pm 0.04	0.053 \pm 0.002	0.051 \pm 0.001	0.062 \pm 0.001
	Obese n=53	4.55 \pm 1.23	34.6 \pm 5.9	0.78 \pm 0.04	0.067 \pm 0.003	0.051 \pm 0.002	0.065 \pm 0.013
Polonsky et al, 1986	Normal n=10	-	-	-	0.057 \pm 0.006	0.054 \pm 0.006	0.06 \pm 0.002
	Diabetic n=7	-	-	-	0.037 \pm 0.004	0.031 \pm 0.004	0.057 \pm 0.002

Equation 4.4 yields an estimated long half-life of 29.2 minutes for newborns (Polonsky et al., 1986). The kinetic parameters were then individually calculated using these cohort specific values, as per (Van Cauter et al., 1992):

$$k_2 = F(b - a) + a \quad (4.5)$$

$$k_3 = \frac{a b}{k_2} \quad (4.6)$$

$$k_1 = a + b - k_2 - k_3 \quad (4.7)$$

where $a = \log(2)/(\text{short half life})$, and $b = \log(2)/(\text{long half life})$. The resulting calculated value for k_3 for all neonates was $k_3 = 0.0644 \text{ min}^{-1}$. This value is within the reported normal clearance rates from other well known studies, shown in Table 4.2.

4.2.4 Trend Analysis

Endogenous insulin secretion was calculated for each sample using Equations 4.3 – 4.7. Results were analysed with respect to patient BW, weight, GA, the DOR, CRIB2 score, maternal cortisol treatments, sex, multiple birth, and ethnicity, to determine if there were any trends within sub cohorts with these characteristics. Insulin secretion was also examined with respect to species or interventions previously reported in several prior studies in the literature to affect insulin secretion (Andronikou and Hanning, 1987, Cser and Milner, 1975, Grasso et al., 1975, Grasso et al., 1990, Grasso et al., 1973, Grasso et al., 1968, Grasso et al., 1970,

Reitano et al., 1978), namely BG, glucose, and protein intake. Linear models were fit using Matlab[®]'s lsqnonlin function.

4.2.5 Statistical Analysis

Results are presented as median (IQR) or number (%) as appropriate. Statistical analyses were carried out in Matlab[®]. Non-parametric data were log transformed to approximate normal distributions and compared using the Student's t-test (ttest2). Binary outcomes, such as survival and multiple births, were tested using the Fishers exact test. The Lilliefors test (lillietest) was used to assess the results of the transformations. P-values < 0.05 were considered statistically significant.

4.3 Results

4.3.1 Insulin Secretion and Clinical Characteristics

Across all samples the median BG and plasma insulin were 7.5 mmol/L and 14.3 mU/L, and the median insulin secretion was 8.2 mU/kg/L/min, as shown in Table 4.3. BG, plasma insulin, plasma C-peptide concentrations and insulin secretion decreased following randomisation (Figure 4.1).

Table 4.3: Insulin secretion and plasma glucose, insulin, IGF-1, C-peptide and cortisol concentrations. Numbers are presented as median [IQR].

Characteristic	Value
Number of Samples	54
Day after randomisation	7 [0 – 14]
BG, mmol/L	7.5 [5.1 – 10.5]
Plasma insulin concentration, mU/L	14.3 [8.5 – 26.2]
Plasma IGF I concentration, nmol/L	0.80 [0.47 – 1.12]
Plasma C-peptide concentration, nmol/L	2.3 [1.1 – 4.2]
Cortisol at randomisation, nmol/L	278.7 [251.1 – 416.6]
Insulin secretion, mU/kg/L/min	8.2 [3.5 – 17.8]

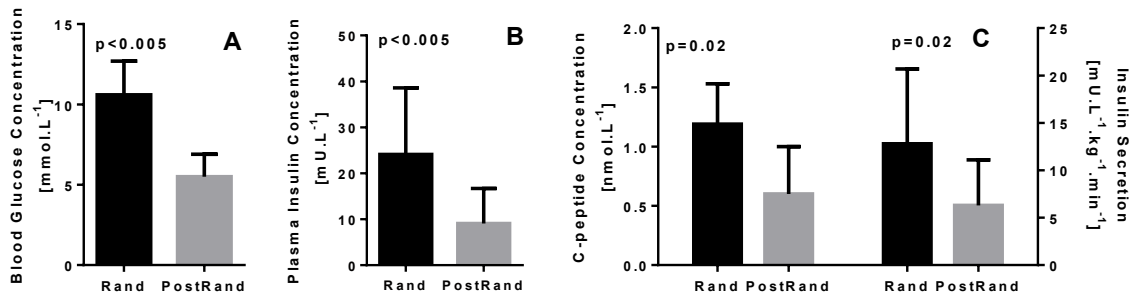


Figure 4.1: Blood glucose, plasma insulin and C-peptide concentrations and insulin secretion at the time of randomisation to the HINT trial and 7-14 days later. Blood glucose (A), plasma insulin (B), and plasma C-Peptide concentrations and insulin secretion (C) from samples taken at randomisation (DOR=1; black), and on days 7 or 14 after randomisation (DOR>1; grey). Data are median (IQR).

There was no difference between TGC and control groups in BG (5.8 [4.8 - 11.0] vs. 8.0 [5.3 - 10.6] mmol/L, $p=0.27$), plasma insulin concentration (14.3 [8.0 - 27.6] vs. 14.3 [8.2 - 27.6] mU/L, $p=0.75$), plasma IGF-1 (0.76 [0.46 - 1.14] vs. 0.93 [0.51 - 1.22] nmol/mL, $p=0.83$), C-peptide concentration (0.51 [0.24 - 1.19] vs. 0.98 [0.42 - 1.56] mmol/L, $p=0.09$), or insulin secretion (6.0 [3.1 - 11.8] vs. 11.1 [3.9 - 18.1] mU/kg/L/min, $p=0.16$). There were no differences in BG, plasma insulin concentrations, and insulin secretion with ethnicity ($p\geq 0.29$). There was not a statistically significant correlation between insulin secretion and CRIB 2 score ($r^2=0.04$, $p=0.33$) or plasma cortisol concentration at randomisation ($r^2=0.05$, $p=0.30$).

Weight at time of sample and insulin secretion were not strongly correlated ($R^2=0.1$). There was no clear trend of insulin secretion with GA. The median insulin secretion tended to be higher in neonates of lower GA at randomisation, or with lower weight, but this group had significantly higher BG than at post-randomisation (DOR>0) (Figure 4.1, $p<0.005$). Separating the cohort into two groups based on gestational age with a cut-off of 26 weeks, gave the greatest significant difference in insulin secretion (GA>26 weeks: 4.9 [3.0 - 9.8], GA \leq 26 weeks: 9.8 [3.8 - 19.9] mU/kg/L/min) based on GA ($p=0.03$), and the IQR is lower for the GA>26 group, showing lower variability.

Weight at time of sample and insulin secretion were not strongly correlated ($R^2=0.1$). There was no clear trend of insulin secretion with GA. The median insulin secretion tended to be higher in neonates of lower gestational age at randomisation, or with lower weight, but this

group had significantly higher BG than at post-randomisation (DOR>0) (Figure 4.1, $p<0.005$). Separating the cohort into two groups based on GA with a cut-off of 26 weeks, gave the greatest significant difference in insulin secretion (GA>26 weeks: 4.9 [3.0 - 9.8], GA \leq 26 weeks: 9.8 [3.8 - 19.9] mU/kg/L/min, $p=0.03$), and the IQR is lower for the GA>26 group, showing lower variability as well.

Of 54 samples, 41 were from singleton births. More babies from non-singleton birth were male (70% vs. 39%), but both groups were similar in GA, weight, PNA and CRIB 2 score than babies from singleton birth ($p\geq 0.69$). BG was significantly higher in the singleton group (8.6 [5.6 - 11.1] vs. 5.5 [4.8 - 7.5] mmol/L, $p=0.03$), but insulin secretion was not significantly different in babies from singleton compared with non-singleton births (9.1 [3.7 - 19.4] vs. 5.1[3.2 - 10.2] mU/kg/L/min, $p=0.28$).

Table 4.4: Clinical characteristics, nutritional data and plasma insulin and c-peptide concentrations in boys and girls. Data are presented as median [IQR] or n(%). PN is parenteral nutrition, and EN is enteral nutrition. TGC is the tight glucose control group. Nutrition data is a daily total for the day the sample was taken.

Characteristic	Sex		
	Boys	Girls	p
n	25	29	-
Randomised to TGC	15 (60)	10 (33)	-
Gestational Age (GA) [wks]	26 [26-27]	26 [25-26.5]	0.98
Weight [g]	930 [750-1043]	815 [700-979]	0.17
Z score	0.03 [-0.76 – 0.11]	0.05 [-0.88 – 0.13]	0.72
SGA	3 (12)	4 (14)	-
Multiple birth	9 (36)	4 (14)	0.06
Post Natal Age (PNA) [d]	9 [3.5-17]	11 [4.5-17]	0.81
Maternal Diabetes	1 (4)	0	1.0
CRIB 2 score	12 [11-14]	12 [10-13]	0.99
Died	4 (16)	2 (7)	0.27
BG [mmol/L]	6.9 [5.5-10.2]	7.6 [4.9-10.8]	0.61
Plasma Insulin [mU/L]	14 [6.8 – 26]	16 [8.9 – 7]	0.30
IGF I [nmol/L]	0.96 [0.54 – 1.31]	0.72 [0.45 – 1.06]	0.33
C-peptide [mmol/L]	0.51 [0.16-0.90]	1.09 [0.46-1.64]	0.005
Insulin Secretion [mU/L/kg/min]	4.7 [2.1-8.3]	11.7 [5.3 – 18.7]	0.003
PN glucose [g/day]	4.6 [0-8.8]	3.6 [0-8.6]	0.34
EN lactose [g/day]	2.1 [0.6-11.9]	6.5 [0.5-14.8]	0.42
Protein intake [g/day]	3.2 [2.5-4.0]	2.8 [2.5-3.8]	0.52
Average dextrose intake [mg/kg/min]	9.4 [7.7-11.1]	9.3 [7.8 – 11.8]	0.79

Insulin secretion was higher in girls (Table 4.4). This difference was seen both at randomisation (18.0 [13.3-27.1] vs. 6.7 [3.5 - 9.4] mU/kg/L/min, $p=0.01$) and post randomisation (8.2 [4.8 - 13.0] vs. 3.6 [1.2 - 7.6] mU/kg/L/min, $p=0.03$). There was no significant difference in insulin secretion between TGC and control groups for either sex (girls: 12.5 [4.9 - 20.4] vs. 11.7 [8.0 - 17.4] mU/kg/L/min, $p=0.81$; boys: 4.7 [1.8 - 7.3] vs. 6.5 [2.2 - 11.5] mU/kg/L/min, $p=0.48$). BG, plasma insulin concentration, nutrition amount or delivery method were not significantly different between the sexes (Table 4, $p\geq 0.34$).

4.3.2 The Effect of Nutrition and Insulin on Insulin secretion

Neither protein intake nor total dextrose intake for the day significantly affected the rate of insulin secretion (Figure 4.2). Adjusting for BG at the time of the sample did not affect this result, with high and low protein intakes being equally scattered with respect to BG and insulin secretion rate.

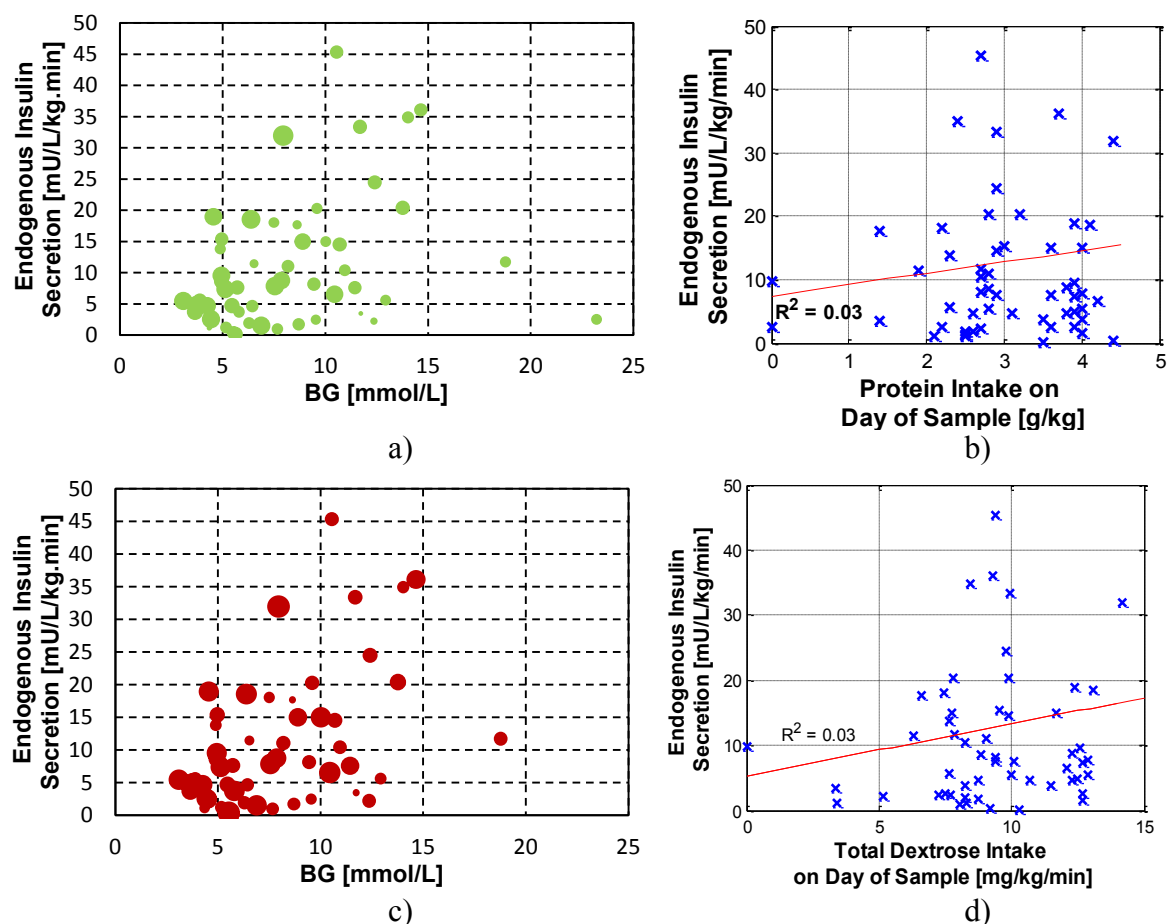


Figure 4.2: Endogenous insulin secretion and blood glucose with respect to protein (a & b) and total dextrose intake (c & d) on the day the sample was taken. In parts a) and c) data points are scaled in size by the magnitude of nutritional intake, with data points from infants with a larger mass of intake being larger in size.

There was a positive and expected relationship between plasma insulin concentration and insulin secretion, but it was weak ($R^2=0.36$), as shown in Figure 4.3b. There was also no clear relationship between both BG and plasma insulin with insulin secretion (Figure 4.3a). Insulin secretion was lower at the time of samples taken during an insulin infusion ($n=13$) than when not on insulin infusion (3.7 [$1.8 - 6.9$] vs. 9.8 [$4.7-17.8$] mU/kg/min, $p=0.02$) with no statistically significant effect on BG (insulin infusion: 6.9 [$5.4 - 9.5$] mmol/L vs. not on insulin: 7.6 [$5.0 - 11.1$] mmol/L, $p=0.46$) or plasma insulin concentration (insulin infusion; 19.9 [$9.2 - 26.7$] mU/L vs. not on insulin; 13.2 [$6.6 - 27.1$] mU/L, $p=0.25$). Thus, none of these effects, as evident in Figure 4.3, had a clear, significant trend.

4.3.3 Insulin Secretion and Blood Glucose Concentration

There was a weak relationship between insulin secretion and BG (Figure 4.4). This relationship was stronger in female babies, with a weak correlation for males. The difference between the sexes in insulin secretion was true over the entire BG range ($p < 0.005$), with no statistically significant difference between clinical characteristics ($p \geq 0.17$), plasma insulin concentration ($p=0.30$), or nutrition regimes ($p \geq 0.34$). Thus, Figure 4.4 also shows separate male and female models for secretion in these cohorts, as well as an overall cohort model.

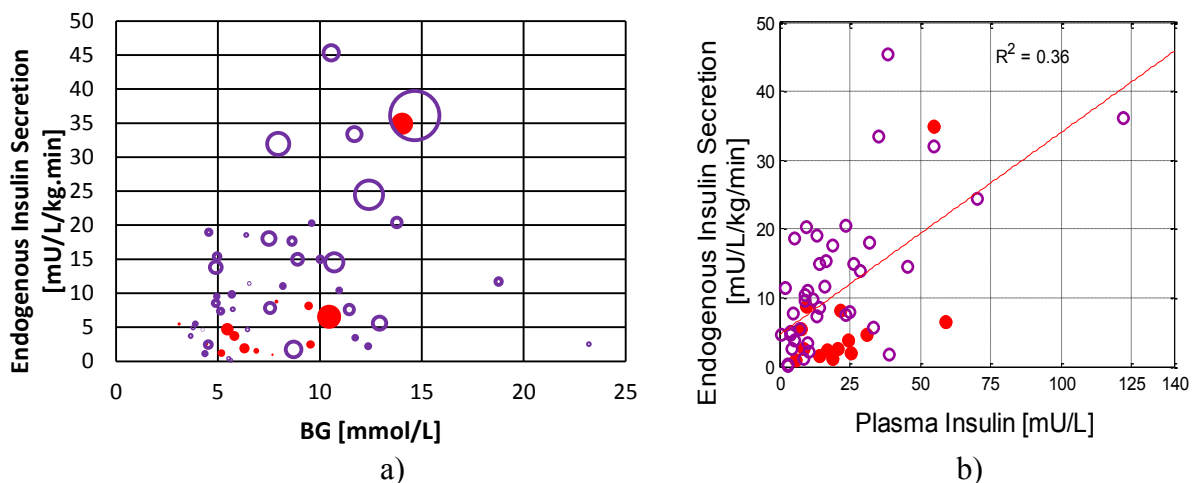


Figure 4.3: Endogenous insulin secretion with a) BG and plasma insulin concentration, and b) plasma insulin. In parts a) data points are scaled in size by the magnitude of plasma insulin, with larger data points representing samples with higher plasma insulin concentration. Open (○) purple and closed (●) red circles denote results from infants not receiving and receiving exogenous insulin respectively at the time of sampling.

These sex based insulin secretion models are defined in Equation 4.8:

$$u_{en} = \begin{cases} \max(4.2, -1.5 + 1.9 * G) & \text{if female} \\ \max(2.2, -0.37 + 0.86 * G) & \text{if male} \end{cases} \quad (4.8)$$

where 4.2 and 2.2 mU/kg/L/min are the lower bounds on insulin secretion in Figure 4.4.

The whole cohort model in Figure 4.4 is defined:

$$u_{en} = \max(3.3, -1.3 + 1.5 * G) \quad (4.9)$$

Multiple linear regression models that accounted for combinations of GA, weight, postnatal age, BG, dextrose intake and exogenous insulin were not able to predict insulin secretion. In particular, weak correlations of $R^2 \leq 0.2$ indicate the small role of these variables. Thus, these models do not benefit from these added clinical variables.

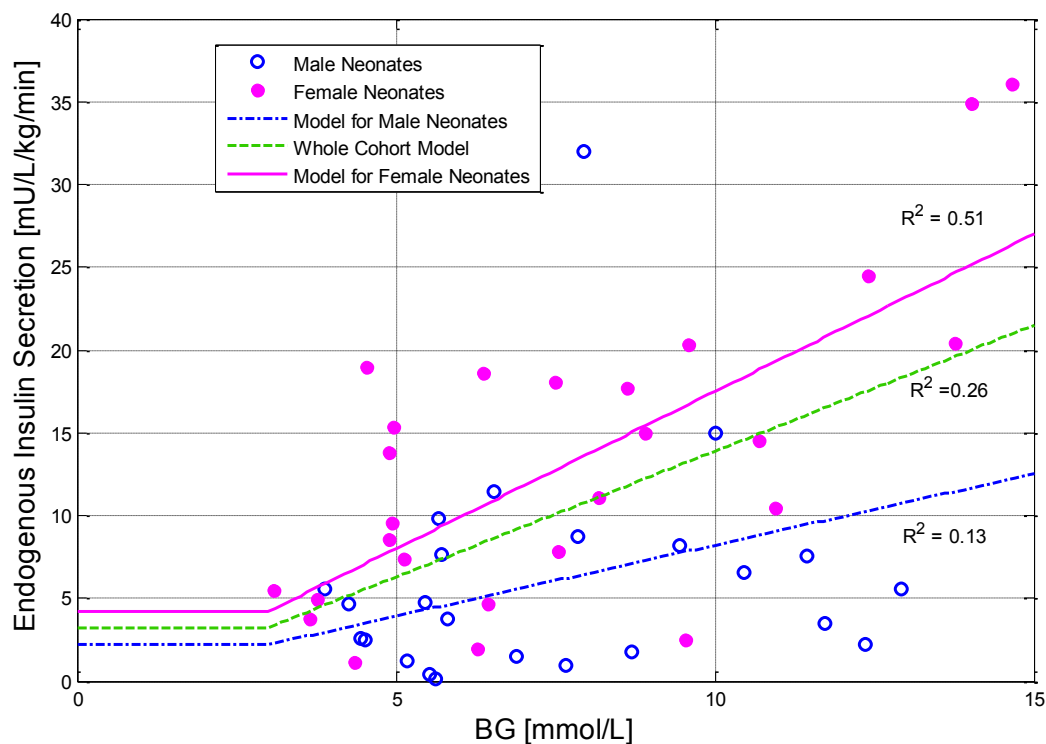


Figure 4.4: Models of endogenous insulin secretion as a function of BG over the whole cohort and male and female sub cohorts. A total of 5 data points from 54 total (9%) were excluded from the analysis as outliers based on a 2-3 fold difference with other data points of similar BG. Some of these data points were from heavier and older patients of GA>29 weeks. However, 2 were from 26 week old male and female neonates.

4.4 Discussion

4.4.1 Insulin Secretion and Sex

Insulin secretion in girls was higher than that in boys, despite similar clinical characteristics, plasma insulin and BG concentrations, and no sex difference in the incidence of neonatal hyperglycaemia in babies eligible for the HINT trial (Alsweiler et al., 2012). The effect of sex on insulin secretion, as measured by C-peptide concentrations, has not been reported previously in preterm babies (Mitanezh-Mokhtari et al., 2004, Sosenko et al., 2007, Tsubahara et al., 2012) or children.

There was a positive relationship between BG concentration and insulin secretion rate. Higher insulin secretion at higher BG concentrations has been reported previously in preterm babies (Adam et al., 1969a) and is supported by research that shows that a reduction in plasma glucose concentration in hyperglycaemic preterm infants is accompanied by a reduction in insulin secretion (Mitanezh-Mokhtari et al., 2004). This correlation was stronger in females, perhaps suggesting that the male sub cohort was sicker or had higher C-peptide clearance. Insulin secretion was similar between males and females at the lower end of the basal BG range, but the linear model fitted to the female sub cohort had a larger slope in Figure 4.4, indicating heightened pancreatic response to changes in BG level. Higher insulin secretion in the females at comparable plasma insulin concentrations could also indicate higher insulin clearances. Glomerular filtration rate (GFR) was not available in these preterm babies, which would have allowed determination of sexual dimorphism in renal clearance of C-peptide. However, GFR has not been observed to be different between the sexes in preterm babies previously (Rhodin et al., 2009).

Differences in insulin concentrations and SI between the sexes have previously been observed during infancy and later in life. Higher plasma insulin concentrations have been observed in girls born at term (Ibanez et al., 2008), and plasma IGF-1 concentrations were also higher in baby girls (Ibanez et al., 2008). Women have been shown to have higher insulin sensitivity (Mathai et al., 2012), and post-pubertal, pre-menopausal women are less likely than men to develop diabetes, although this trend reverses after menopause (Liu and Mauvais-Jarvis, 2010). Potentially, the sex differences in insulin secretion seen in our study could be due to sex hormones, as oestrogen protects against Beta cell apoptosis in mice (Le May et al., 2006) and oestrogen replacement therapy reduces the incidence of Type 2 diabetes in post-menopausal women (Margolis et al., 2004). However in a study looking at age and

sex differences in glucose and insulin metabolism, sex differences in insulin secretion in adults was not observed (Basu et al., 2006).

Preterm birth is associated with higher mortality (Stevenson et al., 2000), and poorer neurodevelopmental outcome (Henderson-Smart et al., 2006) in boys than girls. Animal studies have shown that *in utero* interventions, such as alterations of maternal diet, result in sexual dimorphism of cardiovascular risk factors, such as insulin resistance and raised blood pressure (Aiken and Ozanne, 2013). Recent studies have also shown sexual dimorphism in cardiovascular risk factors after preterm birth in both animal models and human studies (Berry et al., 2013, Feldt et al., 2007, Kaseva et al., 2014). Girls may respond differently to antenatal glucocorticoids, or preterm birth, than boys. Insulin sensitivity is reduced in children born preterm (Hofman et al., 2004) and adults (Dalziel et al., 2005) exposed to antenatal glucocorticoids. Placental 11 β -hydroxysteroid dehydrogenase-2 activity following antenatal betamethasone is greater in girls than boys (Stark et al., 2009), which may reduce the ability of betamethasone to increase insulin sensitivity in girls. As nearly all babies in this study were exposed to antenatal glucocorticoids it was not possible to analyse the effect of antenatal glucocorticoids on insulin secretion. Equally, this consistent exposure also indicated that the results were not biased.

4.4.2 Insulin Secretion and Exogenous Insulin

There was a positive correlation between endogenous insulin secretion and plasma insulin. However, causation and response is hard to separate from this data. Infants in this *post-hoc* analysis were evenly distributed between control and TGC cohorts in the original HINT study (Alsweiler et al., 2012). There are too few data points to conclusively refute or determine models for suppressed insulin secretion in the presence of exogenous insulin, but there was increased insulin secretion in the absence of an exogenous insulin infusion. Male and female infants were evenly spread between the exogenous insulin and no exogenous insulin groups, indicating sex was not responsible for this difference, and vice versa. These data points are tightly clustered with respect to BG, and are thus heavily influenced by one or two points at extremes in BG. In addition, endogenous insulin secretion was not significantly suppressed in the presence of increasing exogenous insulin infusion, as might be expected (Ruotsalainen et al., 1996).

4.4.3 Comparison to Literature

This study determined insulin secretion, calculated from plasma C-Peptide concentrations, in hyperglycaemic preterm babies randomised to tight glycaemic control or standard care. This study used C-peptide measurements, data that relatively few studies have reported for this population (Mitanezh-Mokhtari et al., 2004, Sosenko et al., 2007, Tsubahara et al., 2012). C-peptide concentration is a more accurate indicator of insulin secretion than plasma insulin concentration due to its simple clearance kinetics. In particular, C-peptide is only cleared through the kidney, in contrast to the multiple clearance paths of insulin (Van Cauter et al., 1992).

Surprisingly, no differentiation of insulin secretion rates was seen between differing levels of protein or glucose intake in the neonates, as seen in Figure 4.2. An increase in plasma insulin concentrations in response to a glucose stimulus is well documented in preterm babies (Andronikou and Hanning, 1987, Grasso et al., 1975, Grasso et al., 1990, Grasso et al., 1973, Nakai et al., 1976). In addition, it has been observed that plasma insulin concentrations are higher after a glucose stimulus in the presence of amino acids, such as theophylline theophylline (Cser and Milner, 1975, Grasso et al., 1973, Grasso et al., 1968, Grasso et al., 1970, Salle and Ruiton-Ugliengo, 1977), or glucose priming (Grasso et al., 1975). Potentially, these results reflect the differing physiological stress and degree of prematurity. Some neonates showed high insulin secretion rates with medium-high protein intakes, as would be expected, and it is possible that if a pancreas is compromised due to prematurity, or for any reason, then the presence of protein is unlikely to affect endogenous insulin secretion. Again, a more detailed prospective study would be required to verify this possibility.

There was no statistically significant difference in insulin secretion between babies fed enterally or parenterally, indicating that enteral feeds may not increase insulin secretion in preterm babies. Previous studies have observed higher fasting and post-feed concentrations of glucagon-like peptide-1, which has an incretin effect in term and preterm babies (Padidela et al., 2009), but there are no data available on the effect of this on insulin secretion in preterm babies. In this study, we were unable to further investigate the effect of nutrition on insulin secretion in preterm babies due to the comparison of glucose (parenteral) and lactose (enteral –lactose is broken down into glucose and galactose), and the availability of daily totals of these sugars only. Moreover, babies receiving predominantly enteral feeds were more likely to be older than babies on parenteral feed.

4.4.4 Glucose-Insulin Physiology

The regulation of BG is complex and involves both the liver and the pancreas. Unfortunately, little information is available in this cohort to describe the interactions of these organs and comprehensively describe their contribution to glycaemic regulation. In adults, insulin secretion by the pancreas is regulated by plasma insulin and BG, as well as hormones such as glucagon, cortisol and adrenaline (Bessey and Lowe, 1993, Deibert and DeFronzo, 1980, Gelfand et al., 1984). Stress can also affect secretion (Black et al., 1982), generally through these aforementioned hormones. The variability seen in the secretion dynamics in this neonatal cohort is thus unsurprising when it is considered that their glycaemic response is affected by stress, age (Suneag et al., 1996b), and hormones in the blood, as well as being affected by prematurity.

Multiple linear regression using GA, weight, postnatal age, and exogenous insulin as predictor variables did not significantly alter insulin secretion predictability. The individual effect of each of these factors is impossible to isolate, and very complicated models that could not be specified easily at the bedside, if at all, would be required to successfully model insulin secretion to a higher degree of accuracy. Much of the variability observed in this data can probably be attributed to stress or prematurity, and the data available was insufficient to give an indication of insulin sensitivity between morbidity groups in the cohort.

4.4.5 Limitations

The major assumption required for the calculation of insulin secretion is that of steady state kinetics, an assumption which is considered necessary due to sampling limitations. This assumption limits the studies ability to capture dynamic changes in SI (Bier, 1998). However, it is reasonable given the steady glucose infusion. Importantly, the extremely limited blood volume of extremely preterm babies means it is not possible to take the repeated C-peptide measurements during a glucose tolerance test used in adults to describe dynamic insulin kinetics and secretion with high resolution.

This sampling limitation has two implications. The first is that because detailed information on preterm neonatal metabolism is not currently available, approximations on C-peptide kinetic parameters must be made from adult human data. If the resulting insulin secretion is also calculated using k_3 kinetic values that are approximately 2 standard deviations ($k_3 = [0.05, 0.07]$) from the normal kinetics reported in Table 4.2, and the neonatal cohort falls

within the observed adult range in healthy and unhealthy cohorts, then the insulin secretion could be in error by up to ~20%. However, changing the population constant kinetic parameters does not change any of the results or trends observed, and only shifts these trends. In addition, it is not possible in this cohort to determine patient-specific values. For the purposes of model based control, the k_3 parameter used is thus sufficient. Finally, any scale inaccuracies are absorbed and scale the time varying patient-specific SI parameter (Le Compte et al., 2010a) without significantly altering control outputs.

The second implication is that the insulin secretion models generated in this study are valid in the context of quasi-static clinical conditions. This assumption is reasonable in babies fed predominantly via parenteral infusions that do not tend to change dramatically from hour to hour. In addition, given the limited blood sampling in typical clinical contexts, it is potentially dangerous to try and accurately model what cannot be clinically measured, especially with uncertainty around timing and delivery of feeds and pump based infusions.

4.5 Summary

C-peptide concentrations have been used from blood samples from very preterm neonates in intensive care to estimate steady state endogenous insulin secretion in this cohort, where previous studies have relied on plasma insulin concentrations alone. In this study, it was found that female neonates had a stronger endogenous insulin secretion relationship with BG than their male counterparts, and that overall, BG was the best predictor for endogenous insulin secretion. A linear model of insulin secretion based on BG and sex has thus been created. Insulin secretion was observed to be lower in the presence of exogenous insulin, but data was insufficient to be conclusive. The difference in secretion by sex is a unique result that can impact both control and physiologic understanding of this not well, nor easily studied, cohort.

4.6 Acknowledgements

Dr Jane Alsweiler from the Liggin's Institute, Auckland, New Zealand, for providing the clinical data, and for her clinical insight.

Chapter 5: Modelling Insulin Clearance and Degradation

5.1 Introduction

The previous chapter used a unique set of plasma C-peptide concentrations and well established models of C-peptide dynamics to determine and model insulin secretion in a very premature infant cohort. In this chapter plasma C-peptide and insulin concentrations will be used alongside results from an analysis of data from the literature to determine rate constants for model-based insulin clearance pathways.

Insulin is cleared through a number of pathways, including clearance by the liver, kidneys, and peripheral degradation. In the adult, around 50% of peripheral insulin is removed by the kidneys via glomerular filtration, and proximal tubal re-absorption and degradation (Duckworth et al., 1998). Liver clearance of insulin occurs in two main processes, a first pass hepatic clearance of endogenously secreted insulin and a more general clearance of insulin from circulating blood. Hepatic (liver) insulin clearance is variable and incompletely understood. It involves receptor-mediated processes resulting in saturation of clearance at high insulin concentrations (Duckworth et al., 1998, Duckworth et al., 1988).

Insulin degradation by cells is a complex, receptor-mediated process. Receptor bound insulin can either be released back into the extracellular fluid space or internalised by the cell (Duckworth et al., 1998). Internalised insulin may be returned to the extracellular fluid space or degraded within the cell through several mechanisms. Insulin can also be internalised by a cell via pinocytosis, the pinching of the cell membrane to form a vesicle within the cell. At high insulin concentrations insulin stimulates higher rates of pinocytosis (Duckworth et al., 1998).

Thus, insulin clearance pathways are complex and vary with insulin concentration. Extremely detailed physiological models would be required to capture the specifics of each clearance mechanism. Even in adults, such data is rarely available.

This chapter expands on previous models of insulin kinetics in premature infants (Le Compte et al., 2010a) to model separate insulin clearance pathways. Each pathway has a single clearance parameter associated with it, and saturation effects of receptor-mediated processes are modelled.

5.2 Methods

5.2.1 Modelling Insulin Dynamics

Plasma (I) and interstitial (Q) insulin kinetics in the premature neonate will be modelled with the following equations:

$$\dot{I} = - \frac{n_L I(t)}{1 + \alpha_I I(t)} - n_K I(t) - n_I (I(t) - Q(t)) + \frac{u_{ex}(t)}{V_P m_{body}} + (1 - x_L) u_{en}(t) m_{body} \quad (5.1)$$

$$\dot{Q} = n_I \frac{V_P}{V_Q} (I(t) - Q(t)) - n_C \frac{Q(t)}{1 + \alpha_G Q(t)} \quad (5.2)$$

where n_L (units: 1/min) is hepatic clearance of insulin, α_I (units: L/mU) is the saturation of this clearance, and x_L (unit-less fraction) is the first pass hepatic clearance of insulin secreted into the portal vein by the pancreas. Kidney clearance is determined by n_K (units: min⁻¹), n_I (units: min⁻¹) is the diffusion of insulin between the plasma and interstitial compartments, and n_C (units min⁻¹) is the clearance of insulin via cellular degradation. Insulin degradation by cells is dependent on binding of insulin to insulin receptors, so is saturated by the receptor binding saturation term α_G (units: L/mU). Exogenous insulin delivery (IV infusion or bolus, units mU/min) is $u_{ex}(t)$, and the distribution volumes of insulin in the plasma and interstitial compartments are approximated by plasma and interstitial fluid volumes, V_P and V_Q (units: L/kg) respectively.

Equations 5.1 and 5.2 are structurally similar the ICING model, defined in Chapter 3, Equations 3.7 and 3.8. One difference is that Equation 3.8 has been modified to ensure consistency in the transfer of insulin between compartments with different distribution volumes, as shown in Equation 5.2. These distribution volumes are approximated by the fluid volumes of each compartment, and are now denoted V_P and V_Q . These fluid volumes are in units of L/kg to reflect the significant difference between premature infants of different sizes. Insulin secretion, $u_{en}(t)$ has also been made body mass (m_{body}) dependent and is no longer divided by distribution volume to ensure dimensional consistency with the new secretion model (mU/L/kg/min) developed in Chapter 4.

Saturation of liver clearance of insulin cannot be measured in premature infants, or indirectly determined in this analysis. Thus, the adult value of $\alpha_I = 0.0017 \text{ L/mU}$ is used (Hann et al., 2005, Le Compte et al., 2010a, Lin et al., 2011). In neonates saturation of insulin-mediated

glucose uptake by cells, a receptor-mediated process, has been reported to have little or no saturation in neonates (Farrag et al., 1997). As a result, insulin degradation by cells is also assumed to have no practical saturation ($\alpha_G = 0$).

5.2.2 Clinical Data

The clinical data used in this analysis is the same as was used in Chapter 4, and more details can be found in Section 2.4.1 and Table 4.1. In particular, the plasma C-peptide and insulin concentrations from the clinical blood samples are used alongside results from an analysis of the literature to examine insulin clearances. In this analysis, insulin secretion was directly determined from the C-peptide concentrations according to the analysis in Chapter 4.

5.2.3 Determining Insulin Clearances

Insulin clearance kinetics are determined using the model based method of (Pielmeier et al., 2010a). C-peptide kinetics were defined in Equations 4.1 and 4.2. Assuming the forward and reverse diffusive properties of C-peptide between the two compartments are the same, and with C and Y in concentrations [pmol/L] rather than in absolute amounts [pmol] (as originally presented in (Van Cauter et al., 1992)), then Equations 4.1 and 4.2 in Chapter 4 can be re-written:

$$\frac{dC}{dt} = S - n_{I_{cp}}(C - Y) - n_{k_{cp}}C \quad (5.3)$$

$$\frac{dY}{dt} = n_{I_{cp}} \frac{V_P}{V_Q}(C - Y) \quad (5.4)$$

Here C and Y are concentrations [pmol/L] and the rate constants $n_{I_{cp}}$ and $n_{k_{cp}}$ correspond to parameters in the original Van Cauter model:

$$n_{I_{cp}} = k_1 \quad (5.5)$$

$$n_{k_{cp}} = k_3 \quad (5.6)$$

The rate parameter $n_{I_{cp}}$ denotes the rate transport of C-peptide from the central to the peripheral compartment. The transport of C-peptide into the interstitial compartment is scaled by the difference in volumes of distribution in the central plasma (V_P) and interstitial (V_Q) compartments. The parameter $n_{k_{cp}}$ denotes irreversible removal of C-peptide from central compartment via the kidney.

Since no studies have been performed in preterm or term neonates to determine C-peptide kinetics, adult data and methodology (Polonsky et al., 1986, Van Cauter et al., 1992) was used as a reasonable approximation as the kinetic compartments and their functional physiology are the same. The values for these parameters are given in Table 5.1, and were derived using Equations 4.4 - 4.7. All parameters fall within reported adult ranges (Polonsky et al., 1986, Van Cauter et al., 1992).

If the same steady state conditions are assumed as in Chapter 4, Equations 5.3 and 5.4 simplify to:

$$S = n_{k_{cp}} C = k_3 C \quad (5.7)$$

where Equation 5.7 is the same as Equation 4.3, as expected.

Table 5.1: C-peptide model parameter values from Equations 4.4-4.7

Parameter	Value [1/min]
k_1	0.0478
k_2	0.0516
k_3	0.0644

Assuming similar diffusive properties between insulin and C-peptide, the diffusion constant for insulin between the plasma and interstitial compartments, n_I , can be estimated as the C-peptide transfer rate, n_{ICP} , scaled by molar mass (Pielmeier et al., 2010a):

$$n_I = n_{ICP} \frac{m_c}{m_I} = k_1 \frac{m_c}{m_I} \quad (5.8)$$

where $m_c = 3.02kDa$ and $m_I = 5.8kDa$ are the molecular masses of C-Peptide and Insulin, respectively.

To determine the clearance of insulin in the interstitial compartment, n_c , steady state is assumed and Equation 5.2 re-arranged to yield:

$$n_c = n_I \frac{V_P}{V_Q} \left(\frac{I_{ss}}{Q_{ss}} - 1 \right) \quad (5.9)$$

In adults, studies indicate that the steady state interstitial to plasma insulin ratio (Q_{ss}/I_{ss}) is between 0.4 and 0.6 (Gudbjornsdottir et al., 2003, Sjostrand et al., 2005, Sjostrand et al., 1999, Sjostrand et al., 2000). Therefore, in this study, a ratio of 0.5 is assumed.

If steady state conditions are also applied to Equation 5.1, then the sum of the kidney and liver clearances can be estimated:

$$n_K + n_L \approx \frac{-n_I(I_{ss} - Q_{ss}) + \frac{u_{ex}(t)}{V_P} + (1 - x_L)u_{en}}{I_{ss}} \quad (5.10)$$

First pass insulin clearance by the liver is estimated using the adult value of $x_L = 0.67$ (Lin et al., 2011, Lotz et al., 2008). It is also estimated that 30-80% of insulin in the systemic circulation is removed by the kidneys (Rabkin et al., 1984). Renal clearance of insulin occurs via two pathways, glomerular filtration and absorption from the peritubular capillaries. Glomerular filtration compromises about 60% of total insulin clearance by the kidney (Rabkin et al., 1984).

Creatinine or inulin clearances are two common approaches used to estimate glomerular filtration rate (GFR). The rate of GFR for insulin is approximately 90% that of inulin determined GFR (Maack et al., 1979). As such, total insulin clearance via the kidney can be estimated:

$$n_K = \frac{1}{V_P} \times \frac{0.9 \text{ GFR}}{0.6} \quad (5.11)$$

where GFR is in units of ml/min, and divided by 0.6 to approximate total renal clearance based on the proportion due to GFR. The distribution volume of insulin in the central compartment, V_I , is assumed to be the total volume of blood plasma, V_P . Finally, GFR can be estimated from (Coulthard, 1985):

$$\text{GFR} = 0.45 + 0.24m_{bw} + 0.18A_{PN} \quad (5.12)$$

where m_{bw} the birth is weight, and A_{PN} is the postnatal age in weeks. Using Equations 5.10, and 5.11 and 5.12, kidney and liver clearance of insulin can now be differentiated.

5.2.4 Determination of Fluid Distribution Volumes

Distribution volumes of glucose and insulin have not been determined for premature infants. To approximate these distribution volumes, the distribution volume of insulin in the central compartment is assumed to be the plasma fluid volume (V_P). The distribution volume in the peripheral insulin compartment is assumed to be equal to the interstitial fluid volume (V_Q).

Previously, it was indirectly assumed that interstitial and blood plasma fluid volumes of distribution for insulin were essentially equal (Le Compte et al., 2010a), as in Equations 3.2 and 3.3. This assumption is not necessarily correct for neonates, and a literature search was carried out on PubMed to determine how plasma and interstitial fluid volumes change over time or between patients. As interstitial fluid volume is the extracellular fluid volume (ECV) minus the plasma volume (V_p), extracellular fluid volumes were also included in the search.

Key search terms included: plasma, extracellular, interstitial, fluid volume, and neonate. Exclusion criteria included lack of reported values, or non-human or adult fluid volumes. In premature and term infants, fluid volumes are strongly correlated with weight. Fluid volumes were thus analysed in units of mL/kg, as presented by the studies. Results were plotted against postnatal age and reported standard deviations were included as error bars. Model fitting was carried out in Matlab[®] using the plot fitting toolbox.

5.3 Results

5.3.1 Fluid Volumes

Fluid volumes are well reported in preterm neonates, and 10 studies were found using the given search terms with an average study cohort of GA < 33 weeks and BW < 2kg. Of these, 7 reported ECV values (Bauer et al., 1991, Ekblad et al., 1991, Heimler et al., 1993, Modi et al., 2000a, Shaffer et al., 1987, Shaffer et al., 1991, Shaffer and Meade, 1989b) and 4 reported V_p (Bauer et al., 1991, Cassady, 1966, Leipala et al., 2003, Usher and Lind, 1965). Figure 5.1 shows that ECV drops significantly over the first 1-2 weeks PNA, and after that remains relatively constant. Although a lot of variation is seen between study mean values, most studies fall within one standard deviation of each other.

A power law function ($R^2 = 0.44$) was fitted to capture the rapid decrease in ECV over the first few days and subsequent ECV plateau:

$$ECV = 492 \times PNA^{-0.09} \quad (5.13)$$

where PNA is postnatal age in days.

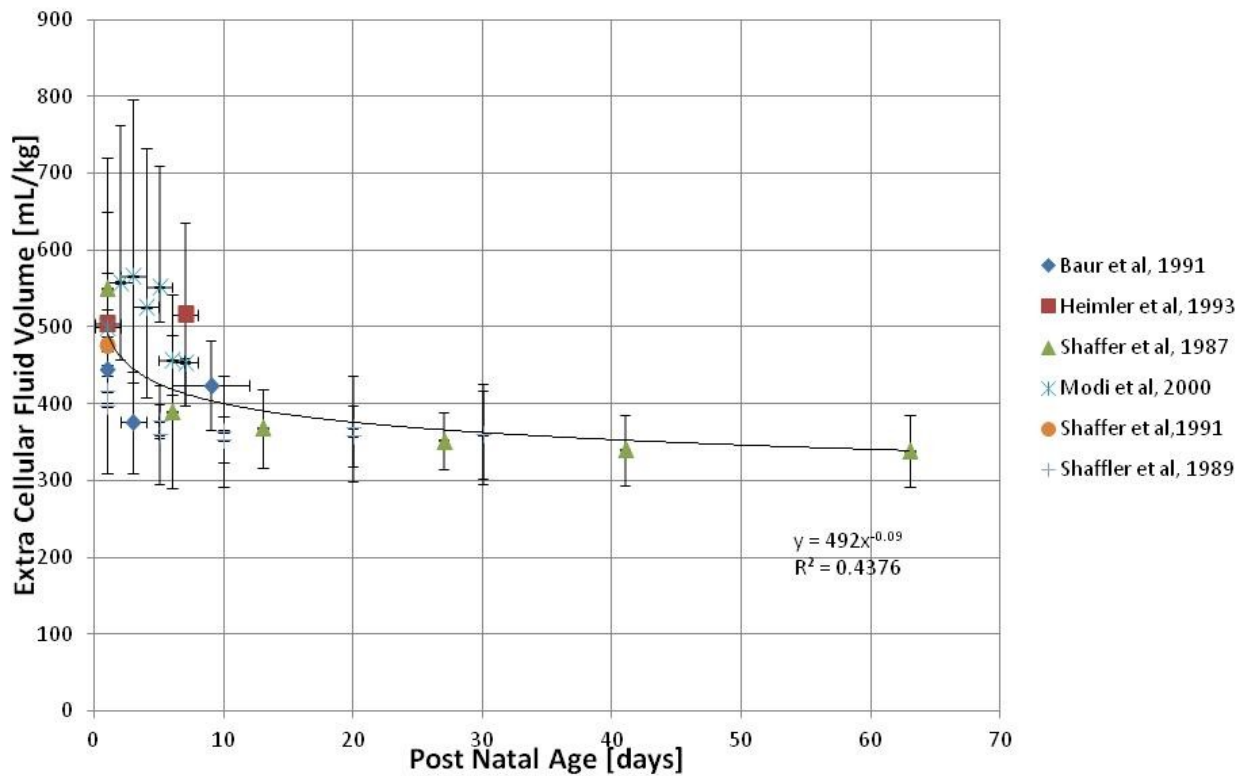


Figure 5.1: Decrease of extracellular volume (ECV) with postnatal age. Vertical Error bars are 1 standard deviation; horizontal error bars cover the range of study days. For the Modi et al. data, the median and range is plotted.

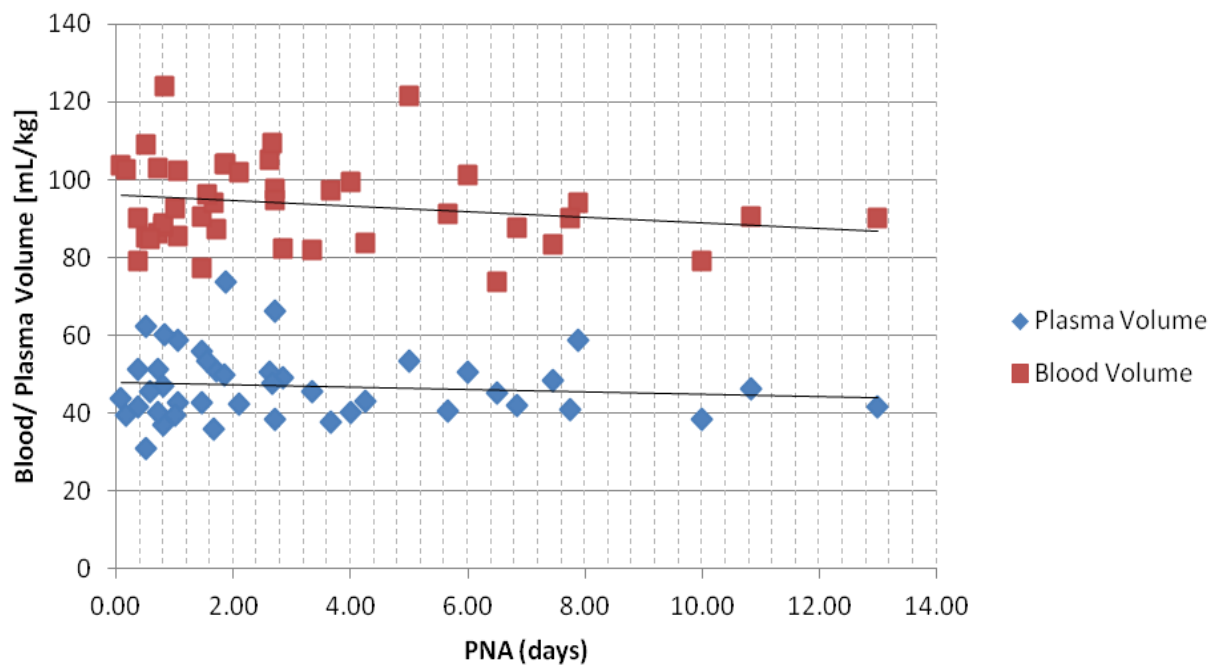


Figure 5.2: Plasma and blood volume changes over time. Data used in this analysis is from patients with BW<2KG, sourced from Usher and Lind (1965).

Figure 5.2 shows that Plasma volume does not significantly change over the first couple of weeks of life. While Ussher and Lind (Usher and Lind, 1965) show a very slight decrease over time ($R^2 = 0.04$), Bauer et al. (1991) shows a slight increase. Therefore, V_P may be assumed to remain essentially constant over time, giving an average plasma volume is 47 mL/kg. The interstitial fluid volume may thus be defined:

$$V_Q = 492 \times PNA^{-0.09} - V_P \quad (5.14)$$

Thus, interstitial fluid volume, with units of mL/kg, can be modelled to decrease with increasing postnatal age.

5.3.2 Insulin Clearance Parameters

The results from the evaluation of Equations 5.8-5.12, using C-peptide data to estimate insulin secretion, are presented in Table 5.2 and Figure 5.3. It should be noted that n_C is scaled by the ratio of plasma volume to interstitial volume (V_P/V_Q). The parameter n_K is made patient specific and depends on a birth weight and postnatal age dependent model of GFR, according to Equation 5.11. The distribution of patient specific values for liver clearance, n_L , is shown in Figure 5.3, with a median clearance of 0.39 min^{-1} .

Table 5.2: Clearance Parameters based on HINT C-peptide analysis and physiological ranges in adult subjects. Unless otherwise stated, results are presented as Median [IQR]

Parameter	Preterm infant value [1/min]	Adult ICING model value [1/min]	Adult range Median [Range] (Lotz et al., 2008) [1/min]
n_I	0.025	0.003	0.28 [0.22-0.36] [L/min] 0.0052 [0.0041 – 0.0119] [1/min]*
n_K	0.021 [0.020-0.22]	0.054	0.15 [0.1-0.21]
n_L	0.39 [0.15-0.70]	0.158	0.06 [0.064-0.53]
$n_c \frac{V_Q}{V_P} = n_I \left(\frac{I_{ss}}{Q_{ss}} - 1 \right)$	0.025	$n_c = n_I$	-

* From Pretty et al. (2014)

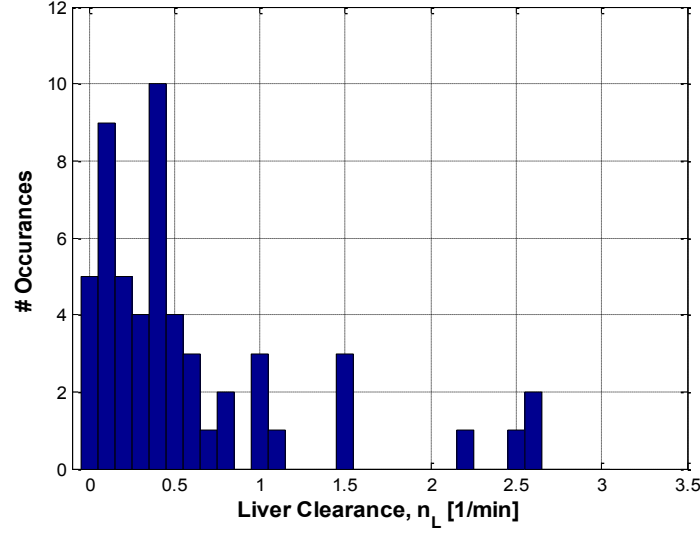


Figure 5.3: Model based liver clearance parameters derived from plasma C-peptide and insulin concentrations.

5.3.3 Summary of Insulin Model

The developed insulin kinetic model is now defined:

$$\dot{I} = -\frac{n_L I(t)}{1 + \alpha_I I(t)} - n_K I(t) - n_I (I(t) - Q(t)) + \frac{u_{ex}(t)}{V_P m_{body}} + (1 - x_L) u_{en}(t) m_{body} \quad (5.15)$$

$$\dot{Q} = n_I \frac{V_P}{V_Q} (I(t) - Q(t)) - n_C \frac{Q(t)}{1 + \alpha_G Q(t)} \quad (5.16)$$

The parameters of the insulin kinetics model presented in Equations 5.1 and 5.2 are defined:

- Liver clearance: $n_L = 0.39$, and $x_L = 0.67$
- Diffusion between compartments: $n_I = 0.025$
- Insulin degradation: $n_C = n_I \frac{V_P}{V_Q} \left(\frac{I_{ss}}{Q_{ss}} - 1 \right)$
- Kidney clearance: $n_K = \frac{1}{V_P} \times \frac{0.9 \text{ GFR}}{0.6}$

Here GFR and fluid volumes are defined:

- Glomerular filtration: $GFR = 0.45 + 0.24 m_{bw} + 0.18 A_{PN}$
- Plasma volumes: $V_P = 0.047 \text{ L/kg}$
- Interstitial fluid volume: $V_Q = 492 \times PNA^{-0.09} - V_P$

5.4 Discussion

This study presents a more physiologically descriptive insulin model for the purposes of glycaemic control. In particular, it focuses on the kinetics of insulin. Previously, the total insulin clearance was modelled by a single clearance parameter in Equation 3.2, determined from literature data and minimisation of model fitting error. Different clearance pathways have been segregated, adding a degradation term to the interstitial compartment, saturable liver clearance, and kidney clearance.

Most of the insulin clearance parameters lie within the reported physiological range for adults (Table 5.2). Insulin diffusion between the plasma and peripheral compartments, n_I , is an order of magnitude faster than the adult ICING model, but is lower than the $n_I = 0.0476 \text{ min}^{-1}$ in adults reported by Lotz et al. (2008). It is higher than the IQR of Pretty et al. (2014), but within the full reported range. Given that the ICING model value for n_I was chosen based on extensive grid search and fitting error minimisation (Lin et al., 2011), and that the value reported by Lotz et al. (2008) was found in fasted adults on low insulin, it is likely that the true physiological value is somewhere in between. That this parameter is slightly lower in premature infants could be explained by prematurity, or reflect assumptions made in this analysis, or both.

Previous work examining insulin transport between blood plasma and the interstitial space in adults has been focused more on determining the mechanism of transport than quantifying transport rate. Trans-capillary insulin diffusion has been identified as one of the rate limiting steps of insulin action (Chiu et al., 2008, Hamilton-Wessler et al., 2002), and is not saturated (Hamilton-Wessler et al., 2002, Steil et al., 1996), suggesting diffusion-based or similar modes of transport. Two studies suggest that insulin transport between plasma and interstitial fluid has a half life of 30-90 minutes (Hamilton-Wessler et al., 2002, Miles et al., 1995), suggesting that the summation of n_I and n_C is between $0.007\text{-}0.023 \text{ min}^{-1}$. There is thus large variability in reported clearance and transport of insulin. This variability could reflect assumptions such as the rate parameter being independent of insulin concentration, or unknown physiological interactions and limitations on insulin transport which impact study design and results.

Birth weight and age based modelling of plasma volumes and GFR allow inter-compartment transport and renal clearances to be patient-appropriate. This approach is in contrast to the adult case, where these volumes are held constant. In addition, during the first week of life

there is significant weight loss in the premature infant as the extracellular fluid compartment contracts. Modelling this contraction will prevent long term bias in identified SI profiles, thus providing more accurate SI estimates.

Insulin degradation is being modelled as a process that is unsaturated at high insulin concentrations. Farrag et al. (1997) observed insulin mediated glucose uptake was not saturated with insulin concentration. Insulin mediated glucose uptake is a receptor-mediated process, and receptor-bound insulin is either released back into the extracellular space, or internalised by the cell (Duckworth et al., 1998). As such, the same saturation parameter is applied to both insulin-mediated glucose uptake and insulin degradation. In this case it has been set to zero, due to the lack of observed saturation of insulin-mediated glucose uptake.

Many of the study limitations are similar to those discussed in Chapter 4. In particular, the intermittent nature of the blood sampling in this extremely fragile cohort means that the assumption of steady state conditions was necessary. Thus, an assumption of approximate clinical steady state conditions underlies both the insulin secretion model, and insulin clearances. This means that the model is less likely to be transiently accurate around times of rapid change, for example large changes in exogenous insulin infusion. Given that insulin infusions change clinically every 3-6 hours, and no exogenous insulin boluses are used, this assumption appears reasonable in this clinical cohort.

5.5 Summary

Clinical data with plasma C-peptide and plasma insulin concentrations was used with an analysis of results reported in the literature to model insulin clearance dynamics. Insulin is cleared by several main pathways, including the liver, the kidneys, and peripheral degradation. Insulin clearance parameters were determined for very premature infants and results were within the range reported in literature. Saturation of liver clearance at high insulin concentration was modelled. Insulin-mediated glucose uptake has been observed to be unsaturated with insulin in premature infants. As this receptor-mediated process is related to insulin degradation, insulin degradation was assumed to be unsaturated.

Distribution volumes of insulin in the plasma and interstitial compartments were assumed to be equal to the fluid volumes of these compartments. The extracellular fluid compartment contracts significantly during the first week of life, while plasma volume remains relatively

constant. Fluid volumes in neonates are reported as mL/kg, and as such the models derived here for plasma and interstitial fluid volumes enable calculation of these fluid volumes in a patient appropriate manner, based on age and weight.

5.6 Acknowledgements

Dr Jane Alsweiler from the Liggin's Institute, Auckland, New Zealand, for providing the clinical data.

Chapter 6: Modelling Endogenous Glucose Production

Chapters 4 and 5 analysed insulin secretion and clearance kinetics. Chapter 6 is the first of two chapters concerned with modelling glucose metabolism. This chapter examines variability around endogenous glucose production (EGP), and attempts to quantify some of this variability through modelling.

6.1 Introduction

EGP is the formation of glucose by the body from substrates. It is a physiological function that normally assists in self-regulation of BG levels and the avoidance of hypoglycaemia. It encapsulates two main metabolic processes: (1) gluconeogenesis, a metabolic pathway generating glucose from non-carbohydrate carbon substrates; and (2) glycogenolysis, by which the body generates glucose through the breakdown of glycogen to glucose.

EGP can be measured by tracer studies (e.g. (Hovorka et al., 2002, Kalhan et al., 1986, Vicini et al., 1997)). However, because of the inherent fragility of the extremely low birth weight (ELBW) cohort, the true fasting rate of EGP cannot be explicitly determined experimentally in this way. This introduces significant uncertainty to metabolic models that rely on this value. Studies measuring unfasted EGP are relatively few for this cohort, and in general. What data there is in the literature for similar cohorts must be extrapolated. In addition, inter-patient variability has led to significant variation between results and conclusions in these studies.

Studies have attempted to quantify the variability of EGP, but with mixed results. Gluconeogenesis has been shown to persist in premature infants receiving total parenteral nutrition (TPN) (Chacko and Suneag, 2010). EGP has been shown to remain unaffected by amino acid administration (Diderholm et al., 1999, Poindexter et al., 2001, Van Kempen et al., 2003a), but lipids have been shown to support (Suneag, 2003a) or enhance (Van Kempen et al., 2006) EGP. Glycerol has been shown to enhance gluconeogenesis (Suneag, 2003b) and to be a principle gluconeogenetic substrate (Suneag et al., 1996b, Suneag et al., 1999b). Furthermore, the extremely preterm infant is capable of generating glycerol at a rate similar to much larger, full term infants (Suneag et al., 1996b, Suneag et al., 1993). Preterm infants have been shown to produce glucose at a rate similar to (Suneag et al.,

1996b, Sunehag et al., 1993) or exceeding (Chacko and Sunehag, 2010, Keshen et al., 1997, Tyrala et al., 1994) that of full term infants, or adults. There is also evidence of some relationship between GA and EGP (Chacko and Sunehag, 2010) and/or weight and EGP (Sunehag, 2003a). Overall, EGP is hard to measure and poorly understood in this premature infant cohort.

6.2 Methods

The analysis in this chapter utilises both literature review and clinical data. A literature review was carried out to examine reported values and trends for EGP. From identified trends, EGP models were created, and their efficacy analysed with respect to model fit to clinical data and control outcomes in simulation. EGP population constants based on literature distributions were also used to examine the effect of EGP on control in a clinical patient cohort, across the entire range of possible EGP values. These methods are summarised in Figure 6.1, and the overall goal is to find a best fit and realistic model.

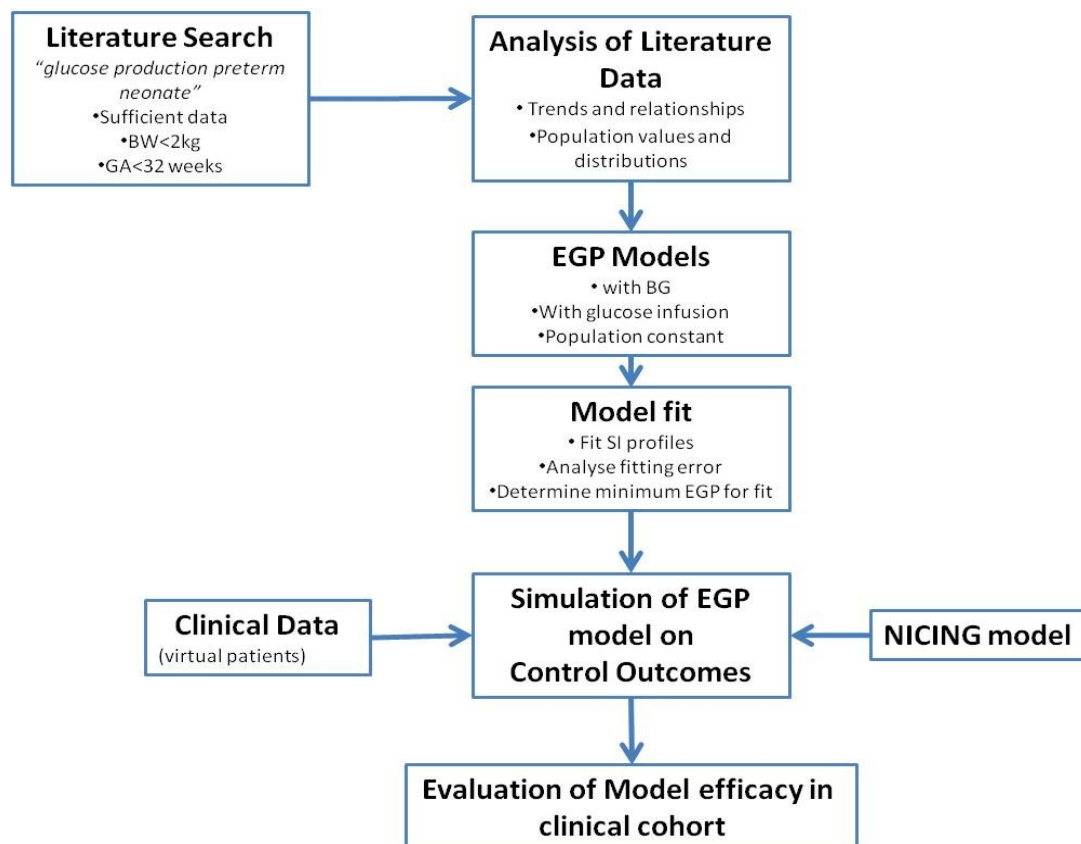


Figure 6.1: Methodological approach to analysis, modelling, analysis, and evaluation of models of EGP in very premature infants.

To evaluate the best model in the context of glycaemic control in the VLBW cohort, two approaches were used. The first was fit to clinical data, which evaluates the ability of a given model to capture clinically observed BG dynamics. The second was control performance, which evaluated the clinical safety of models with similar fit to clinical data using virtual trials. The second approach is required because, within physiological thresholds, EGP and SI trade off in the model. The second approach indirectly minimises SI variability and balances this minimisation with a focus on choosing an EGP value or model for safety in control.

6.2.1 Literature Review

A literature search was carried out using search criteria of “glucose”, “production”, “preterm”, and “neonate” in the PubMed database. Studies were excluded if they were associated with maternal or foetal diabetes, subjects were not human, subjects were full term or older, or studies were unrelated to glucose metabolism. Studies were chosen from the remaining literature on the basis that they reported sufficient data, including the rate of EGP, BG, BW < 2 kg, GA < 32 weeks, and glucose infusion (GI) feed regimes. A total of 177 data points were collected from 21 studies. Study methods and primary conclusions are summarised in Table 6.1, and cohort demographics are summarised in Table 6.2. Of these studies, 7 (33%) also reported plasma glucagon, and 8 (38%) reported plasma insulin.

EGP was analysed for trends with respect to BW, GA, GI and BG, as well as glucagon and plasma insulin where available. A linear function was then fit using least squares for EGP versus GI, the strongest correlated pair of parameters. A piecewise linear function was also used to describe the suppression of EGP with increasing BG. The piecewise linear model was chosen because of high inter-patient variability in the data.

6.2.2 Model Fit to Clinical Data

The clinical patient cohort outlined in Table 6.2, consists of data from 21 retrospective patients (25 patient episodes, 3098 hours, 943 BG measurements) and 40 patients (54 patient episodes, 5014 hours, 1632 BG measurements) from prospective BG control studies in neonatal intensive care using STAR (Le Compte et al., 2009, Le Compte et al., 2012). Patients who received no insulin, or had no BG measurements, for more than 8 hours between treatments were separated into multiple patient episodes. Patient episodes were separated by more than 24 hours, and were at least 4 BG measures in length.

Table 6.1 Overview of literature studies in endogenous glucose production in premature infants

Study	Methods		Study Conclusion(s)
	Tracers	Tracer Infusion duration	
Hertz et al. (1993)	[6-6-d ₂] glucose	Primer dose: 5min Study period 4 hours	<i>Clinically stable, extremely premature infants suppress glucose production and increase glucose utilization in response to increased glucose infusion. Increasing the rate of glucose delivery results in no change in whole body proteolysis in these infants.</i>
Sunehag et al. (1993)	[6-6-d ₂] glucose	Primer dose: 10min Study period: 2 hours	<i>Infants born at GA <28 weeks have a capacity to produce glucose on their 1st day of life at rates close to or even exceeding those reported in term infants.</i>
Sunehag et al. (1994b)	[6-6-d ₂] glucose	Primer dose: 5min Study length: 160-180 min	<i>Very immature newborn infants have an incomplete and varying capacity to respond to glucose infusion with suppression of glucose production. Insulin seems to be more important than plasma glucose in the regulation of glucose homeostasis in these infants.</i>
Tyrala et al. (1994)	[6-6-d ₂] glucose	Primer dose: 1min Study length: 2 hours	<i>Infants who weigh <1100g utilise 3-4 times more glucose per kg of body weight than adults, reflecting their higher brain to body weight ratio. EGP provided only ~ 3rd of the glucose required</i>
Sunehag et al. (1996b)	[6-6-d ₂] glucose, [2- ¹³ C] glycerol	Primed 2 hours	<i>Extremely preterm infants are capable of generating glycerol at a rate within the range reported for term and near term newborns. The infants were also capable of converting part of this glycerol to glucose, providing a contribution to hepatic glucose production comparable to that found in more mature newborns.</i>
Keshen et al. (1997)	[U- ¹³ C] glucose	4 hour	<i>Neonates whose birth weights are less than 1200g have a particularly high glucose production rate, secondary to enhanced gluconeogenesis.</i>
Sunehag et al. (1997)	[U- ¹³ C] glucose [2- ¹³ C] glycerol	10 hours.	<i>During TPN gluconeogenesis accounts for 1/4 - 1/3 of glucose rate of appearance after 8-10 Hrs of reduced glucose supply.</i>
Sunehag et al. (1998)	[U- ¹³ C] glucose [2- ¹³ C] glycerol	8 hours	<i>In VLBW infants receiving TPN, gluconeogenesis is maintained equally well by endogenous or exogenous amino acid supply.</i>
Diderholm et al. (1999)	[6-6-d ₂] glucose [2- ¹³ C] glycerol	3.5 hours	<i>No significant change in plasma glycerol concentrations, glycerol production, and the fraction of glycerol converted to glucose after theophylline administration. The percentage of glucose derived from glycerol increased.</i>
Sunehag et al. (1999b)	[U- ¹³ C] glucose [2- ¹³ C] glycerol [6-6-d ₂] glucose	11 hours	<i>VLBW infants receiving TPN, normoglycemia was maintained during reduced glucose infusion by glucose production primarily derived from gluconeogenesis. Glycerol was the principal gluconeogenic substrate.</i>
Farrag and Cowett (2000)	N/A	N/A	<i>Adult-like response to insulin requires maturation beyond the neonatal period. Ontogeny of glucose utilisation responsiveness to insulin occurs before that of glucose production.</i>

... Table 6.1 continued

Study	Methods		Study Conclusion(s)
	Tracers	Tracer Infusion duration	
Poindexter et al. (2001)	[6-6-d ₂] glucose + unlabeled glucose	3 hour study	<i>In response to amino acid infusion rates of endogenous glucose production were unchanged.</i>
Sunehag (2003b)	[U- ¹³ C] glucose	8 hours	<i>In very premature infants, parenteral glycerol enhances gluconeogenesis and attenuates time dependent decrease in glucose production.</i>
Sunehag (2003a)	[2- ¹³ C] glycerol	10 hours	<i>In parenterally fed very premature infants, lipids play a primary role in supporting gluconeogenesis.</i>
Van Kempen et al. (2003b)	[6-6-d ₂] glucose [2- ¹³ C] glycerol	6 hours	<i>Preterm infants can only partly compensate a decline in exogenous glucose supply by increasing endogenous glucose production rate. The ability to maintain the plasma glucose concentration after a decrease in exogenous supply is better preserved in infants >30 wk owing to more efficient adaptation of peripheral glucose utilization.</i>
Van Kempen et al. (2003a)	[6-6-d ₂] glucose [2- ¹³ C] glycerol	6 hours baseline (unlabeled) infusion Study length: 3 hours	<i>Administration of alanine does not stimulate gluconeogenesis in preterm infants.</i>
van Kempen et al. (2005b)	[6-6-d ₂] glucose [2- ¹³ C] glycerol	6 hours baseline Study length: 1hr	<i>Increase in glucose production after glucagon was similar in appropriate for GA (AGA) and small for GA (SGA) infants, and mainly due to an increase in glycogenolysis. Based on the assumption that glycogenolysis is an indicator of liver glycogen content, this data does not support the hypothesis that liver glycogen content is lower in preterm SGA compared to AGA infants after the first postnatal day.</i>
van den Akker et al. (2006)	[U- ¹³ C] glucose	Study length: 6-7 hours	<i>The anabolic state resulting from amino acid infusion in the immediate postnatal period resulted from increased protein synthesis and not decreased proteolysis. Energy required for additional protein synthesis was not derived from increased glucose oxidation</i>
Van Kempen et al. (2006)	[6-6-d ₂] glucose [2- ¹³ C] glycerol	9 hours	<i>Intralipid enhanced glucose production by increasing gluconeogenesis in preterm infants.</i>
Chacko and Sunehag (2010)	[6-6-d ₂] glucose	Primer ² H ₂ O Study Period: 8 Hours	<i>Gluconeogenesis is sustained in preterm infants receiving routine TPN providing glucose at rates exceeding normal infant glucose turnover rates and accounts for the major part of residual glucose production. Gluconeogenesis is not affected by the glucose infusion rate or blood glucose concentration.</i>
Chacko et al. (2011)	[U- ¹³ C] glucose	11 hours	<i>In extremely low birth weight infants receiving total parenteral nutrition, GNG is a continuous process that is not affected by infusion rates of glucose or concentrations of glucose or insulin.</i>

Table 6.2: Summary of patient metrics between the literature data and the clinical data cohorts

Cohort	Cohort Medians		
	Gestational Ages [weeks]	Birth Weight [g]	Blood Glucose [mmol/L]
Hertz et al. (1993)	25.5	8900	6.1
Sunehag et al. (1993)	26	796	3.5
Sunehag et al. (1994b)	27	1196	4.8
Tyrala et al. (1994)	Range: 23-28	Mean: 858	5.7
Sunehag et al. (1996b)	26	865	3.1
Keshen et al. (1997)	28	1110	6.5
Sunehag et al. (1997)	Mean: 27	Mean: 1020	Mean 3.1
Sunehag et al. (1998)	27.6	1060	3.8
Diderholm et al. (1999)	28.5	1160	3.8
Sunehag et al. (1999b)	27	1050	2.8
Farrag and Cowett (2000)	-	677	3.9
Poindexter et al. (2001)	32	1500	4.8
Sunehag (2003b)	28	1030	3.5
Sunehag (2003a)	27.5	995	3.0
Van Kempen et al. (2003b)	29.1	1140	3.9
Van Kempen et al. (2003a)	<32	AGA	3.9
van Kempen et al. (2005b)	30.5	1244	4.6
van den Akker et al. (2006)	27.4	946	5.4
Van Kempen et al. (2006)	29.4	1335	4.2
Chacko and Sunehag (2010)	Mean: 26.5	Mean :955	Mean: 8.9
Chacko et al. (2011)	25.4	820	4.1
Median literature data [IQR]	27.5 [26-29]	1080 [921-1315]	4.1 [3.8 - 5.4]
Median clinical data [IQR]	26.3 [25.0-27.8]	848 [636-940]	6.9 [6.2-7.9]
P-value	< 0.0001	< 0.0001	< 0.0001

Virtual patients were created from clinical data through the identification of model-based S_I profiles (See section 9.2). Once the S_I profile was identified, model solutions were generated for G , I and Q (Matlab[®], ode45). The model used to fit to clinical data was the insulin kinetics model developed in Chapters 4 and 5, with the BG model of Equation 3.1:

$$\dot{G} = -p_G G(t) - S_I G(t) \frac{Q(t)}{1 + \alpha_G Q(t)} + \frac{P_{TPN}(t) + P_{EN}(t)}{V_{g,frac}(t)m_{body}} + \frac{EGP m_{body} - CNS m_{brain}}{V_{g,frac}(t)m_{body}} \quad (6.1)$$

Parameter values for the population constants were from Le Compte et al (Le Compte et al., 2010a), and are summarised in Table 6.3.

Accuracy of model fit to clinical data was one metric used to evaluate the effect of the new EGP models. This fitting error is defined as the average percentage difference between the real and modelled BG levels at BG measurements. In addition, when using an integral based fitting method (Hann et al., 2005), the identified S_I must remain positive to be physiologically correct; the lower limit was set to $S_I = 1 \times 10^{-7}$ L/mU/min.

Where fitting error was poor, with modelled BG failing to reach clinical measurements, a negative S_I had been forced to a lower limit of 1×10^{-7} L/mU/min. In such cases, Equation 6.1 was re-arranged and EGP was identified for a perfect BG fit using $S_I = 1 \times 10^{-7}$ L/mU/min. The resulting EGP_{\min} values gave an indication of the magnitude of minimum EGP required to adequately fit clinical data under the assumption of minimum peripheral SI. These results should thus show the minimum level of inter-patient variability in EGP as it uses a minimal S_I and only extreme cases seen by larger fitting errors. Thus, it is likely that differences in EGP were larger, but cannot be identified without further data that is currently unavailable.

6.2.3 Control Based Analysis

Generated models of EGP, as well as EGP as a population constant, were examined through simulation of glycaemic control using virtual S_I profiles generated by the model fit process. The range of constant EGP values used can be found in Table 6.4, and represents the wide range of likely EGP values observed in the literature. Percentiles of literature reported EGP were investigated, and other specific and relevant EGP values between the median and 95th percentile were included for completeness.

Table 6.3: Glucose model parameters used in the EGP analysis

Parameter	Value
p_G	0.003 min^{-1}
α_G	0
$V_{g,frac}(t)$	GA based, as per Avery et al. (1994)
CNS	$0.088 \text{ mmol/kg/min}$
m_{brain}	14% of m_{body} [kg]

For each EGP model, S_I profiles were identified for each patient (Section 9.3), and whole cohort stochastic models were generated (Le Compte et al., 2010b). Control was tested using clinically validated virtual trial methods (Chase et al., 2010a, Le Compte et al., 2009) and the control protocol simply selected insulin such that the predicted outcome likelihood of $BG > 4.4$ mmol/L was greater than or equal to 5%, as per the basic protocol in the adult ICU (Fisk et al., 2012). A limit of 0.03 U/kg/hr was applied on the increase in insulin dose, as was applied by Le Compte et al (Le Compte et al., 2009). Control performance was measured by the percentage time in band (BG between 4.0-8.0 mmol/L), while the number of patients experiencing severe hypoglycaemic events ($BG < 2.6$ mmol/L) evaluated safety.

Table 6.4: Values of constant EGP used to investigate effect on control

Percentile	EGP [mg/kg/min]
5 th	0.29
25 th	1.40
50 th	2.1
75 th	4.6
95 th	7.7
Currently used [19]:	5.11
Other EGP:	5.50, 6.00, 6.50, 7.00, and 7.50

6.3 Results

6.3.1 EGP and Patient Metrics

Comparing reported values of EGP with BW and GA showed little or no correlation in Figure 6.2. High inter-patient variability between similar patients across these studies is seen in the large scatter of EGP across all metrics. Similarly, as shown in Figure 6.3, there is no distinct correlation between BG and EGP, or plasma insulin and EGP. Across all literature studies, a suppression of EGP with increasing GI can be seen in Figure 6.3. However, at any given GI rate there is still significant variation in EGP, with no clear distribution with BW or GA. The piecewise linear trend of GI and EGP shown in Figure 6.3 is defined:

$$EGP(GI) = \begin{cases} -0.55 \times GI + 4.96, & 0 < GI \leq 7 \\ 1.11, & GI > 7 \end{cases} \quad (6.2)$$

Where EGP and GI have units of mg/kg/min.

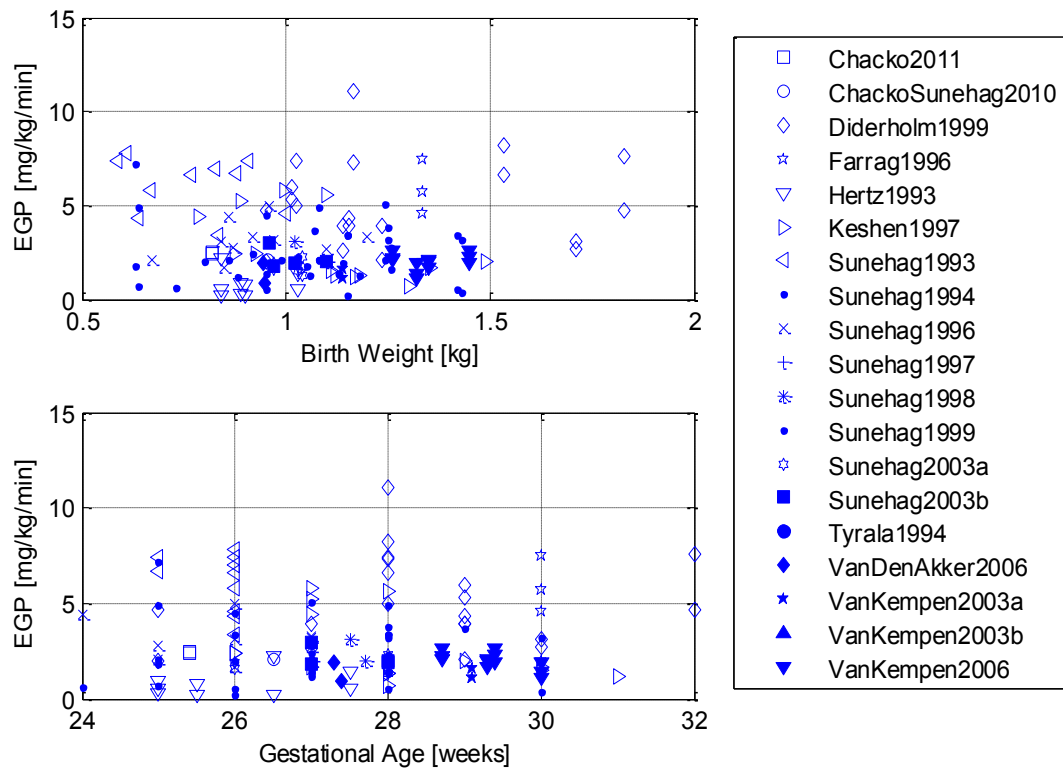


Figure 6.2: Relationships between endogenous glucose production (EGP) with birth weight (BW) and gestational age (GA).

6.3.2 EGP and Glucagon

Results in Figure 6.4 show large variation in EGP for any given glucagon concentration, based on data from (Chacko et al., 2011, Cowett et al., 1983, Diderholm et al., 2007, Helander and Fandriks, 2014, Suneag et al., 1994b, Van Kempen et al., 2003b). At low glucagon concentrations EGP is higher in neonates born at $GA \geq 29$ weeks, and seems to decrease with increasing glucagon concentration. For neonates born at $GA < 29$ weeks, EGP is approximately constant across the entire plasma glucagon range.

For any given plasma insulin concentration, plasma glucagon shows large variation, particularly with lower plasma insulin levels, as seen in Figure 6.5. There is no distinguishable and consistent trend in plasma glucagon levels with age, birth weight (not shown), or plasma insulin. Plasma glucagon levels drop with increasing blood glucose concentration in Figure 6.5a, as might be normally expected. In addition, plasma glucagon levels are seen to be higher with higher gestational age, with the same trend of decreasing plasma glucagon with increasing BG. As there was no clear relationship between glucagon and EGP, no further models were developed and tested in control.

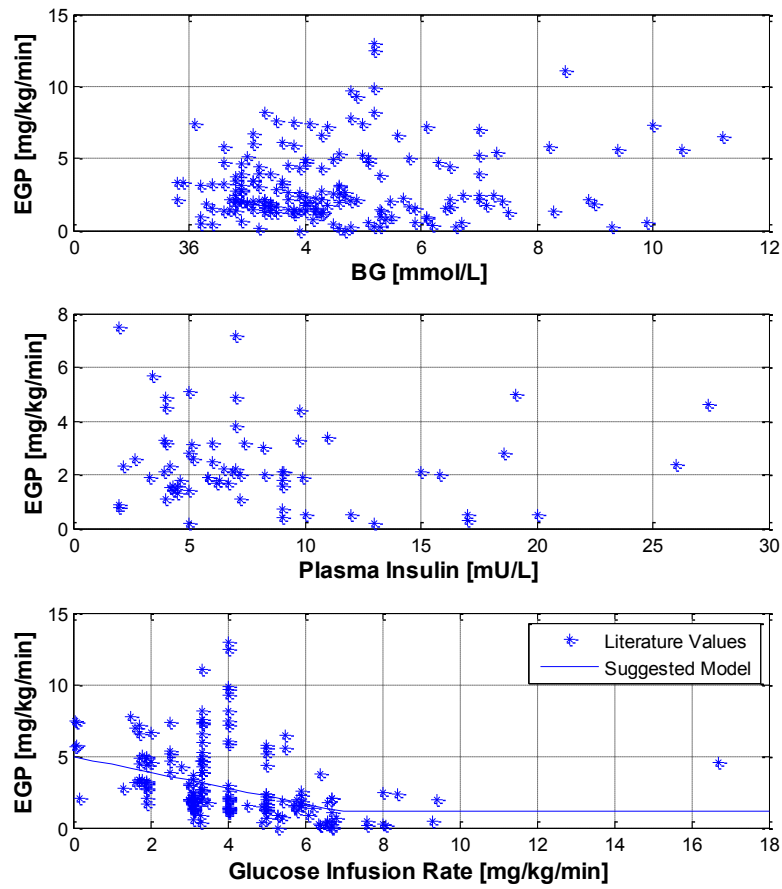


Figure 6.3: Relationships between endogenous glucose production (EGP) with blood glucose (BG), plasma insulin, and glucose infusion rate (GI).

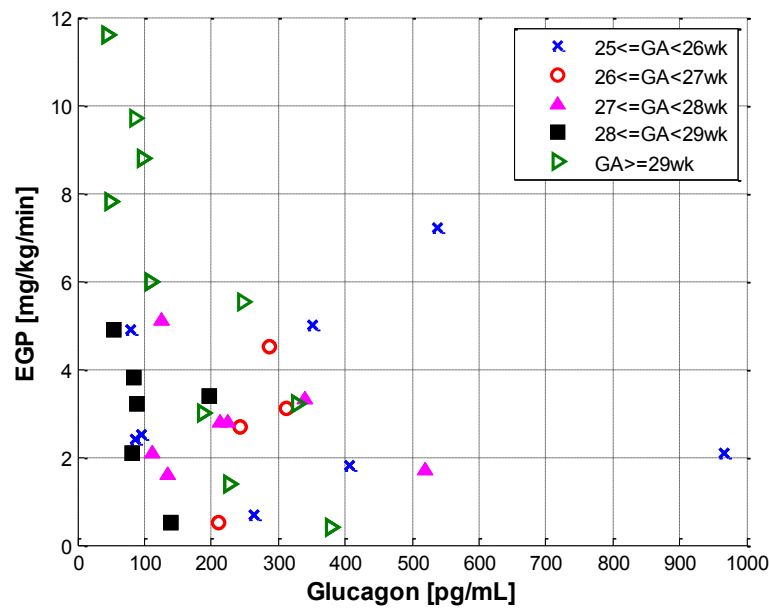


Figure 6.4: Endogenous glucose production (EGP) and plasma glucagon concentration. Based on data from (Chacko et al., 2011, Cowett et al., 1983, Diderholm et al., 2007, Helander and Fandriks, 2014, Sunehag et al., 1994b, Van Kempen et al., 2003b).

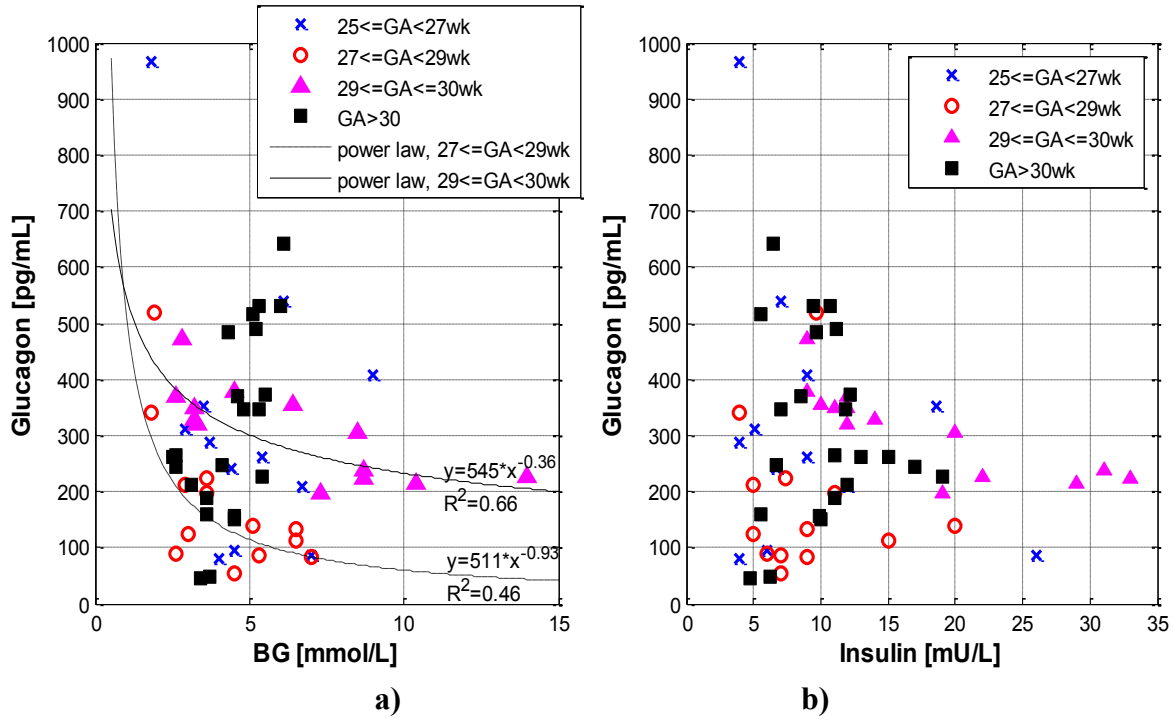


Figure 6.5: Plasma glucagon and: a) blood glucose (BG) and b) plasma insulin concentration. Based on data from (Borgstrom et al., 1957, Chacko et al., 2011, Cowett et al., 1983, Diderholm et al., 2007, Heimler et al., 1993, O'Neill, 2003, Sunehag et al., 1994a, Sunehag et al., 1994b, Tian et al., 2012)

6.3.3 EGP and Blood Glucose

Across all studies, EGP and BG are plotted in Figure 6.6, and show high variability across the glycaemic range. However, a sub-cohort of studies shows some degree of increase in EGP with BG, indicative of dysfunction due to stress response. These studies are plotted in Figure 6.7a, with each study showing a different trend in the magnitude of EGP with respect to BG. All of these specific studies show high variation, as reflected in the R^2 values from 0.2-0.5. If all the remaining studies are considered, a contrasting trend of suppression of EGP with increasing BG can be seen. This suppression exists to varying degrees among and between studies, as shown in Figure 6.7b.

From Figure 6.7b a suppressed EGP with BG can be modelled. The EGP variation with BG is modelled as:

$$EGP(BG) = \begin{cases} 4, & BG < 2 \\ 5.75 - 0.88 * BG, & 2 \leq BG \leq 6 \\ 0.5, & BG > 6 \end{cases} \quad (6.3)$$

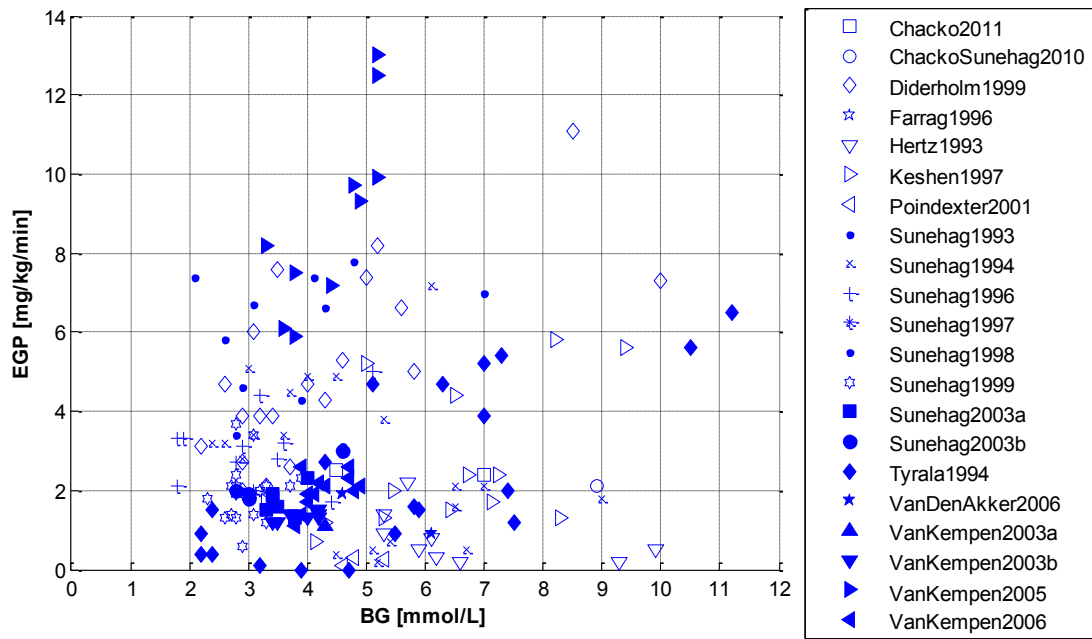


Figure 6.6: Endogenous glucose production (EGP) as a function of blood glucose (BG) over the literature cohort of 177 data points.

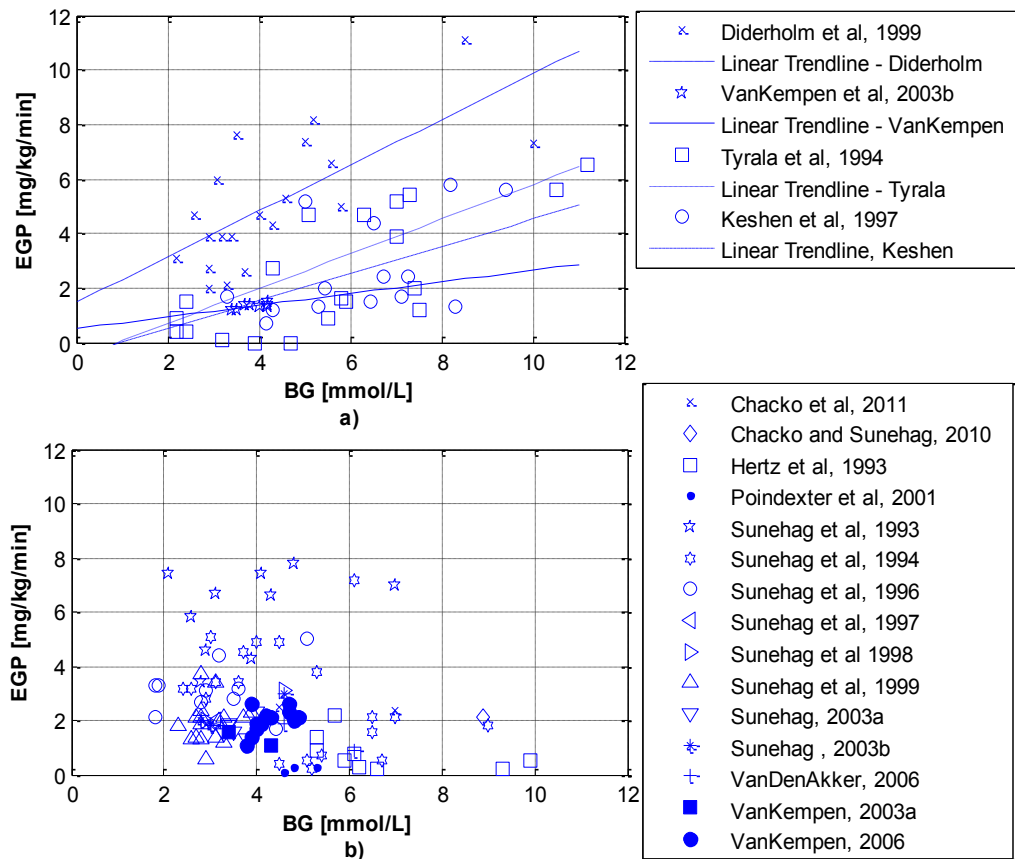


Figure 6.7: Sub-cohorts of studies which show: a) increasing endogenous glucose production (EGP) with blood glucose (BG), and b) suppression of EGP with BG.

EGP has units of mg/kg/min and BG mmol/L. With the suppression of EGP with BG, there was a fitting error of 3.57% over the whole cohort. However, many patients had one or more instances where S_I was constrained to a lower limit of $S_I = 1 \times 10^{-7}$ L/mU/min without fitting the data, indicating insufficient EGP production in the model.

To estimate patient specific EGP over these periods, the EGP was reverse calculated using an assumption of an absolute minimum $S_I = 1 \times 10^{-7}$ L/mU/min, giving EGP_{\min} in Figure 6.8. This is the lowest EGP required to fit the data assuming minimal insulin-mediated glucose removal. These minimum values suggest that EGP in hyperglycaemic infants is generally higher than the literature data for slightly heavier and non-hyperglycaemic (Table 6.2) infants, and is not suppressed by elevated BG, as might be expected in a stress response. EGP_{\min} is also scattered across the cohort with a far wider spread than the literature data, indicating greater inter- and intra- patient variability than previously reported with no clear trend.

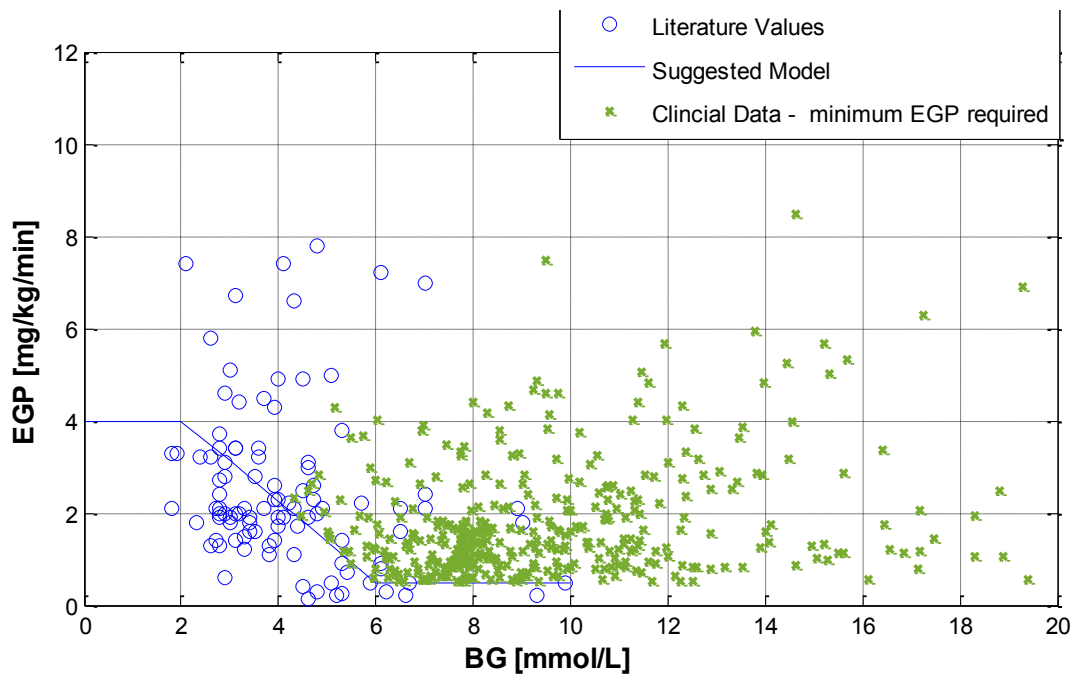


Figure 6.8: Comparison of literature based model of endogenous glucose production (EGP) with blood glucose (BG) and the minimum EGP required for adequate fit to clinical data (EGP_{\min}).

Table 6.5: Comparison of model fit and control performance across different endogenous glucose production (EGP) models.

Case	Fitting	Control			
EGP [mg/kg/min]	Fitting error %	Time in band (4.0 - 8.0 mmol/L)	Hyperglycaemic (BG > 10 mmol/L)	Mild hypoglycaemic (BG < 4.0 mmol/L)	Severe hypoglycaemic # BG < 2.6 mmol/L
Suppressed EGP(BG)	3.57	EGP too low to sustain modelled BG levels, indicating inability of the EGP model to replicate clinical results.			
0.3 (5th)	4.26				
1.40 (25th)	2.93				
2.1 (50th)	2.64	66.7	13.4	1.2	2
4.20	2.40	69.3	12.6	1.0	2
4.6 (75th)	2.39	69.8	12.5	1.0	2
5.11*	2.38	70.8	11.7	1.0	1
5.50	2.38	70.3	11.9	1.1	1
6.00	2.37	70.8	11.6	1.1	1
6.50	2.37	70.8	11.7	1.2	1
7.00	2.37	70.8	11.7	1.2	1
7.50	2.36	71.4	11.4	1.2	0
7.7 (95th)	2.36	71.3	11.6	1.2	1
EGP(GI(t))	2.58	67.1	13.2	1.0	2

* Currently in use with EGP = 5.11 mg/kg/min

6.3.4 Control Analysis of EGP

The fitting and virtual trial control performance metric results for different EGP models are shown in Table 6.5. EGP as a function of GI and EGP as a function of BG both performed worse than the currently used constant EGP = 5.11 mg/kg/min (Le Compte et al., 2010a). In the first case, fitting error was increased, and in both cases the number of patients with hypoglycaemic events increased.

Due to high variation in EGP in Figures 6.6 and 6.7, a range of constant EGP values were investigated. Table 6.5 shows that EGP values below 2 mg/kg/min had high fitting error due to insufficient EGP to reach clinically measured BG levels, and during simulation EGP was

insufficient to maintain a positive BG. Increasing EGP decreased fitting error and increased the performance and safety of STAR model based glycaemic control. However, all average fitting errors for $EGP > 2 \text{ mg/kg/min}$ are within measurement error of 1-2% for blood gas analysers. Thus, compared to BG values in Figure 6.6, the current value of $EGP = 5.11 \text{ mg/kg/min}$ appears reasonable. From a control standpoint, increasing EGP beyond this value increases control, with $EGP = 6.0 \text{ mg/kg/min}$ providing a best compromise between a likely physiological value and improved control. Beyond this value, the small percentage improvements in Table 6.5 are unlikely to be clinically significant.

6.4 Discussion

6.4.1 Study Results

The piecewise linear models of suppressed EGP with increasing BG shown in Figure 6.8, and EGP with GI, shown in Figure 6.3, resulted in poor fitting and control performance. These poor results are primarily because EGP values in the literature were too low during hyperglycaemia to sustain the BG values measured clinically in the data used in this research. They suggest that hyperglycaemic ELBW premature infants are more greatly stressed than the cohorts studied in the literature. This assumption matches the statistical differences in Table 6.2. Hence, they often fail to suppress or otherwise regulate EGP with BG or GI, compared to normal infants.

In particular, literature data and EGP_{\min} data points did not overlap, as shown in Figure 6.8. The BG data from the literature studies in Table 6.2 and Figure 6.8 were in the normal range. Thus, as noted, it is likely that the majority of these infants were healthy, and therefore representative of normal EGP dynamics. In contrast, the clinical data is based on hyperglycaemic infants in the NICU, with higher average BG levels, suggesting this cohort is less healthy. This result implies that EGP may be higher in these preterm and hyperglycaemic infants, which is physiologically intuitive as BG is likely high, at least in part, due to elevated or unsuppressed EGP due to stress of their condition. These differences mimic the adult ICU situation (Capes et al., 2000, McCowen et al., 2001), which would again suggest that hyperglycaemic infants have less ability to suppress EGP with high BG or GI.

In support of these outcomes, the clinical data patients are typically younger (lower GA), lighter (lower BW), and generally start hyperglycaemic, unlike the literature data. Clinical

data had a median starting BG of 9.8 [IQR: 8.3–12.3] mmol/L, compared to the literature median BG of 4.1 [IQR: 3.8–5.4] mmol/L. These statistics, summarised in Table 6.2, reflect a limitation in the use of literature data to describe EGP model in hyperglycaemic ELBW infants. However, no other data for EGP in this or any similar cohort exists, due to the practical difficulty of measuring EGP in this cohort. Thus, the literature data provides the only valid basis for extrapolation, despite its limitations.

Figure 6.7a suggests that some neonates are at higher BG levels because of a physiological inability to regulate EGP. However, the regulation of BG is overall a complex function of EGP, endogenous insulin secretion, insulin therapy, and nutritional treatment. Thus, it is extremely difficult to define a direct cause and effect relationship between BG and a single variable EGP. This difficulty is partially reflected in the low R^2 values in Figure 6.7a. Higher EGP with increased BG, as seen in the sub-cohort of studies in Figure 6.7a, was not modelled as this model formulation created a positive feedback system within the simulation software, which inhibited the controller's ability to regulate BG to a target band. It was also not reflective of a majority of the literature, or the presumed stress state behaviour of the specific ELBW cohort examined here.

While glucagon was seen to decrease with increasing BG, as would be expected physiologically, EGP also decreased with increasing glucagon, which is counter to known or expected physiological response. As a result, further models considering glucagon were not created or tested in control. This outcome potentially suggests that while the pancreas is able to suppress glucagon production with elevated BG to some degree, the liver is not yet able to respond fully to the EGP stimulus.

As the overall result of this study, it can be concluded that a population constant of $EGP = 6.0$ mg/kg/min is adequate for use in model-based control. This population constant model best accounts for, and reflects, uncertainty due to variability between patients. Increases in controller performance seen in virtual trials using higher assumed EGP values are unlikely to be clinically significant.

6.4.2 Comparison to Literature

No strong relationship was found in a range of literature reports between EGP and BW, plasma insulin, or GA. Keshen et al. (1997) have reported decreasing EGP with increasing body weight in babies less than 31 weeks GA, and with a PNA of 4-9 days. However, this

single result remains unconfirmed by any other study in the literature examined, and was contradicted by the findings of Chacko and Sunehag (2010) in a study with a similar patient cohort. Van Kempen et al. (2003b) reported that the ability of neonates to maintain basal BG levels with a decrease in exogenous insulin is greater in neonates older than 30 weeks GA. However, no studies have specifically investigated EGP over a range of GA, although preterm infant EGP has been shown to be similar or exceed that of term infants (Sunehag et al., 1996a, Sunehag et al., 1993).

Some studies conclude that glucose production has been regulated by BG levels (Hertz et al., 1993, Kalhan et al., 1986), but the majority report the reverse (Chacko et al., 2011, Chacko and Sunehag, 2010, Cowett et al., 1983, van Goudoever et al., 1993). Preterm infants display varying ability to suppress EGP with increasing glucose infusion, with complete (Hertz et al., 1993), incomplete (Sunehag et al., 1994b, Van Kempen et al., 2003b), and failed (Chacko and Sunehag, 2010) suppression reported. Although one study suggests that insulin plays an important role in EGP regulation (Sunehag et al., 1994b), other studies show that EGP is not suppressed by plasma insulin levels (Chacko et al., 2011, Chacko and Sunehag, 2010, Sunehag et al., 1993, van Goudoever et al., 1993). Hence, there is significant variability in fundamental physiological responses between EGP in these main hormonal and demographic characteristics.

6.4.3 Study Limitations

A study in adults by Tigas et al. (2002) suggests that using an isotope infusion period of 5 hours or longer can reduce error in EGP measurements by at least 80% due to the time required for isotopes and substrates to equilibrate. As a result, some of the studies undertaken before 2002, where infusion periods tended to be shorter than 5 hours, may have inherent error in EGP. However, using only literature reported results with an infusion period of 5 hours or greater (Table 6.1) (Chacko et al., 2011, Chacko and Sunehag, 2010, Sunehag et al., 1997, Sunehag et al., 1999a, Sunehag, 2003b, Sunehag, 2003a, Sunehag et al., 1998, van den Akker et al., 2006, Van Kempen et al., 2003b, Van Kempen et al., 2006) changed none of the trends seen for EGP, and did little or nothing to reduce the variation in EGP seen across all metrics. Thus, for the data available, this issue was not a factor.

Similarly to EGP, CNS glucose uptake is also a population constant based on literature data (See Equation 6.1). It is possible that CNS in this population is lower, which would be reflected in this study as a higher EGP compared to the value of $CNS = 0.088 \text{ mmol/kg/min}$

(15.8 mg/kg/min) assumed. In addition, a higher EGP term could result from a need for reduced endogenous insulin production. Hence, the values here are relative to these parameters and assumptions.

Finally, this analysis is only relevant in the context of this model. The same model framework and *in-silico* control modelling approach has been validated in adult cases where much more independent data is available (Chase et al., 2010a). Thus, it is felt that the overall results showing enhanced EGP with elevated BG are realistic and valid, especially given the limited data available or possible for this particular cohort.

6.5 Summary

A wide range of literature studies have been found that report EGP in the very premature cohort. The studies themselves are divided and often contradictory in their conclusions, and no definitive relationship between EGP and BG, plasma insulin, or patient metrics, such as weight and GA, exists. Over all studies, EGP was shown to be highly variable between patients and studies. One consistent trend was that EGP was seen to decrease with increasing glucose infusion over all the literature studies examined. Additionally, two trends were seen with glucose production and BG. The first saw higher EGP at higher BG, and the second saw suppression of glucose production at higher BG. Both of these latter trends are physiologically reasonable. However, given all this variability, STAR glycaemic control was still found to perform best when EGP was modelled as a population constant, with $EGP = 6.0$ mg/kg/min providing a good balance between control performance and the likely physiological range. Finally, a model-based analysis indicated that hyperglycaemic ELBW infants produce glucose at a higher rate than healthy counterparts.

6.6 Acknowledgements

The author would like to acknowledge the contribution of J.N Hewett of the AIC10 final year project team, who assisted in the initial literature search and first analysis of results.

Chapter 7: Modelling Non Insulin-Mediated Central Nervous Glucose Uptake

7.1 Introduction

Glucose utilisation in the body has insulin dependent and insulin independent pathways. Cellular uptake of glucose is achieved via 5 types of glucose transporters (GLUT), of which only GLUT4, which achieves glucose uptake in muscle and adipose tissue, is insulin dependent. The GLUT 1 transporter is found in all cells, but plays a significant role in allowing rapid diffusion of glucose into the brain. The rate of glucose uptake by the brain is relatively constant (Gruetter et al., 1998). The GLUT2 transport is mainly found in liver and pancreatic cells, and plays a role in cellular monitoring of glucose concentrations (Tirone and Brunicardi, 2001, Triplitt, 2012). GLUT3 mediates uni-directional glucose and sodium uptake in neurons, kidneys, and the intestine (Tirone and Brunicardi, 2001, Triplitt, 2012).

Non-insulin mediated cellular glucose uptake in this thesis is simplified to central nervous system (CNS) uptake and other uptake. This approach lumps the effects of GLUT 2 and GLUT3 into one parameter, p_G in Equation 6.1. This chapter will focus on CNS uptake and, in particular, its variation with brain mass.

7.2 Methods

7.2.1 Central Nervous System Uptake

Only two studies in premature infants have directly examined cerebral glucose metabolism in premature infants (Kinnala et al., 1996, Powers et al., 1998). The median value of the study by (Powers et al., 1998) was used as a population constant by Le Compte et al. (Le Compte, 2009, Le Compte et al., 2010a). The study by (Kinnala et al., 1996) examines cerebral metabolism in older premature infants (32-56+ weeks since conception), and is thus beyond the scope of this study as significant development and changes will have occurred over that period. As the CNS parameter directly trades off with EGP in the model, the sensitivity of this parameter and its effect on control has been indirectly examined in the previous chapter.

This analysis will instead focus on the modification of the weight based CNS glucose uptake constant, which has units of mmol/kg/min, through improved modelling and estimation of brain mass.

7.2.2 Other Non-Insulin Mediated Glucose Uptake

In Equation 6.1, the expression $-p_G G(t)$ is used to capture non insulin mediated glucose removal via the kidneys, intestine, pancreas, and liver. As the insulin-mediated glucose uptake expression, $-S_I G Q$, is structurally similar to this non-insulin mediated glucose uptake expression, both patient specific p_G and S_I cannot be uniquely determined at the same time (Hann et al., 2005). The p_G term is assumed to be adequately modelled by the adult value of $p_G = 0.003 \text{ min}^{-1}$, given a lack of non-insulin mediated glucose data specific to premature neonatal physiology.

7.2.3 Brain Mass Estimation

Two methods of estimation of brain mass are examined here. The first treats brain mass as a fixed proportion of body weight, and the second uses head circumference.

Proportion of body mass

Previous work by Le Compte et al utilised brain mass as constant proportion of body mass (Le Compte et al., 2008, Le Compte, 2009). This proportion was estimated using data from Ho et al. (1981), where body and brain mass is reported over a range of preterm infant sub cohorts defined by sex and ethnicity. Ethnicity was defined simply as 'black' and 'white', with

no other details provided. Sub-Cohort characteristics and brain-to-body mass ratios are given in Table 7.1.

Table 7.1: Mean statistics on body and brain mass. Data sourced from Ho et al. (1981)

Ethnicity	Sex	Gestational Age (wk)	m_{body} (g)	m_{brain} (g)	m_{brain}/m_{body} (g/g)
Black	Female	27.3	958	139	0.145
	Male	28.4	1151	157	0.136
White	Female	29.2	1258	165	0.131
	Male	30.4	1434	190	0.132

A brain-to-body mass ratio of 0.140 was determined from the reported results for the cohort with the lowest GA (GA 27 - 28 weeks, ethnicity 'black'). This value is reflective of the ratio of 0.136 obtained if the entire cohort is used. As the hyperglycaemic neonatal cohort seen in previous work has lower gestational age and weight than that of Ho et al. (1981), it is likely that this ratio is slightly higher than 0.14. The body mass based method, denoted the BM model, for brain mass estimation is thus defined:

$$m_{brain} = 0.14 m_{body} \quad (7.1)$$

Head circumference and brain mass

Head circumference may be a more accurate indicator of brain mass than body mass alone, given that brain-to-body mass ratio changes with GA (Dobbing and Sands, 1973) (Table 7.2). A model relating head circumference to brain mass has been proposed by (Cooke et al., 1977). The model was derived from post-mortem analysis of 485 premature infants of GA 18-43 weeks. Small-for-gestational-age (SGA) infants are accounted for using a different parameter set to those that were appropriate-for-gestational-age (AGA). This separation was the result of SGA infants having a statistically higher brain mass for their GA than AGA infants (Cooke et al., 1977).

The brain mass and head circumference model from (Cooke et al., 1977), denoted here as the HC model, is defined:

$$m_{brain} = C^b \times k \quad (7.2)$$

where C is the head circumference in centimetres and m_{brain} is the brain mass in grams. The remaining parameters given are in Table 7.2.

Table 7.2: Parameters in HC model. Data sourced from (Cooke et al., 1977)

Cohort	b	k
Appropriate-for-Gestational-Age	3.001	0.0093
Small-for-Gestational-Age	3.225	0.0048

7.2.4 Patient Data

Glycaemic control records from 88 patients enrolled in the HINT glycaemic control trial (Alsweiler et al., 2012), previously detailed in Section 4.21, were used to generate virtual patients. Patient episodes were created from infants who received insulin therapy and had 5 or more consecutive BG measurements, with a maximum of 8 hours between measurements. A patient episode was not included if no head circumference measurements were taken during the episode, or within 24 hours before the start of an episode.

A total of 48 patients were selected, totalling 82 episodes, and patient demographics are given in Table 7.3. The 82 virtual patient episodes constituted 7032h of care, with 1881 BG measurements (mean 3.7 h/intervention). None of the mothers of patients in the study experienced maternal diabetes. Of the 48 patients, 17 experienced intra-ventricular haemorrhage (IVH), all of whom were AGA infants. IVH was rated on a scale of 1–5 in order of increasing severity, where the 17 patients had a median [IQR] IVH score of 2 [1–3.5] (Alsweiler et al., 2012).

Detailed information about insulin treatments and BG measurements were recorded. Nutritional input was both parenteral and enteral. Parenteral sources of glucose included TPN, dextrose infusions, and dextrose within medications. Enteral feeds were started as early as possible in this trial, and consisted of formula and/or breast milk, depending on breast milk availability. Nutrition was recorded as daily totals, so feed timing and changes in nutrition within a 24 hour period were not detailed. It was thus assumed that the daily glucose total was delivered as a constant parenteral and/or enteral infusion over 24 hours, which is acceptable given this analysis concerns the constant model value, CNS.

Table 7.3: Median [IQR] cohort demographics

Characteristic	Value
Birth Weight (g)	750 [678–894]
GA at birth (weeks)	25.0 [24.0–26.5]
Age at start of trial (days)	4 [2–7]
Ethnicity (%)	
NZ European	28
Māori	35
Pacific Islander	6
Asian	10

SGA infants were defined as patients at or below the 10th percentile of weight for their GA using growth charts from New South Wales, Australia (Beeby et al., 1996) that are used in New Zealand NICUs. Of the entire cohort, 5 of 48 patients were SGA, corresponding to 7 of 82 patient episodes. In addition, 4 of the 5 SGA patients were asymmetric SGA, as determined by the Ponderal index (Vik et al., 1997). Due to the small SGA cohort, asymmetric and symmetric patients are not analysed separately.

7.2.5 Analyses

As the true brain mass could not be measured, the different methods for estimating brain mass were first compared against each other, and then analysed using a control based analysis, similar to that in Chapter 6. SI was fitted from clinical data (See section 9.2), creating virtual patients, using both brain mass estimation models. Glucose and insulin kinetics were modelled using the insulin kinetics model developed in Chapters 4 and 5, and the population constant for EGP determined in Chapter 6. BG model parameters are summarised in Table 7.4. For each brain mass model a stochastic model forecasting SI was created (Le Compte et al., 2010b, Lin et al., 2006).

Table 7.4: Glucose model parameters used in the EGP analysis

Parameter	Value
p_G	0.003 [min ⁻¹]
α_G	0
$V_{g,frac}(t)$	GA based, as per (Avery et al., 1994)
CNS	0.088 mmol/kg/min (15.8 kg/kg/min)
EGP	0.033 mmol/kg/min (6.0 mg/kg/min)

Virtual trials were used to assess the sensitivity of control based outcomes to brain mass estimation methods. The control protocol used was that detailed in Section 6.2.3. Measurement interval was set to 4 hours. As the 'true' brain mass is unknown, four simulation cases are considered, and these are detailed in Table 7.5. Comparison of control outcomes is made using nonparametric statistics. Distributed data are compared using the Wilcoxon rank-sum (also known as the Mann–Whitney U test: Matlab[®], ranksum). Values of $p < 0.05$ are considered statistically significant.

Table 7.5: Combinations of assumed model and controller-perceived model used in virtual trials

		S_I profile in Control	
		HC-fit control	BM-fit control
S_I profile in Simulation	HC-fit virtual profile	A. Assume HC gives the best estimate, and control with it.	B. Assume HC gives the best estimate, but control with BM
	BM-fit virtual profile	C. Assume m_{body} gives the best estimate, but control with HC.	D. Assume BM gives the best estimate, and control with it.

7.3 Results

7.3.1 Brain Mass Model Comparison

The median [IQR] brain mass estimated using the BM model, based on a body mass was 112 [973 – 129] g. In comparison, the median [IQR] brain mass estimated using the HC model, based on head circumference, was 121 [102 – 144] g. Estimations of brain mass from the BM and HC models are compared in a Bland–Altman plot in Figure 7.1. The figure shows a bias towards the BM model estimating a significantly lower value for m_{brain} ($p < 0.005$). The median difference ($m_{brain} (BM) - m_{brain} (HC)$) was -9.8 g, with an IQR of $[-19.6, 0.9]$ g.

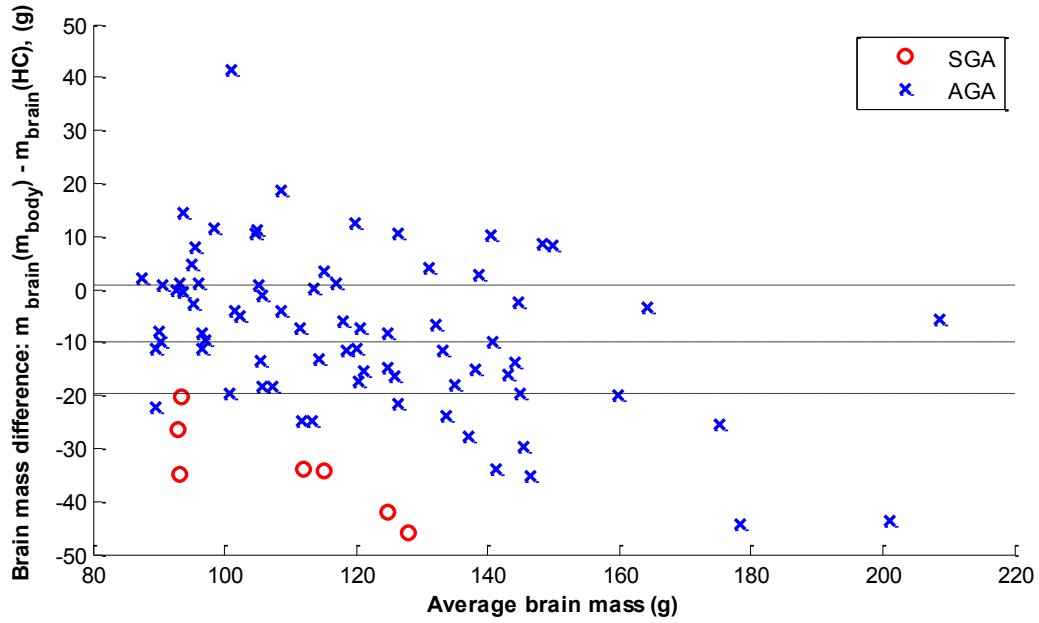


Figure 7.1: Comparison of BM and HC model estimations for brain mass, highlighting SGA infants.

The bias towards a much lower estimation of brain mass via body mass proportion is stronger in the SGA patients. These SGA infants have a median difference of -34g , with an IQR of $[-40, -28]\text{ g}$ ($p < 0.001$). As a result, the distinction between AGA and SGA infants that Cooke et al. (1977) required for their HC model may also be necessary when using a body mass estimate.

In Figure 7.2 the data points from Figure 7.1 are further differentiated by their mass for GA percentile bands. The downward bias is present across all centiles and average brain masses, indicating that m_{body} estimations of brain mass tend to be lower than HC brain mass estimations. This result indicates that the body mass and head circumference methods represent significantly different measures of brain mass.

7.3.2 Effect on Model-Based Insulin Sensitivity

Patients are fit to the NICING model to generate SI profiles for virtual patients using both models for brain mass estimation. The resulting SI profiles identified are summarised in Table 7.6. The median SI value is 1.1% lower using the BM model. However, this difference is not significant ($p=0.47$).

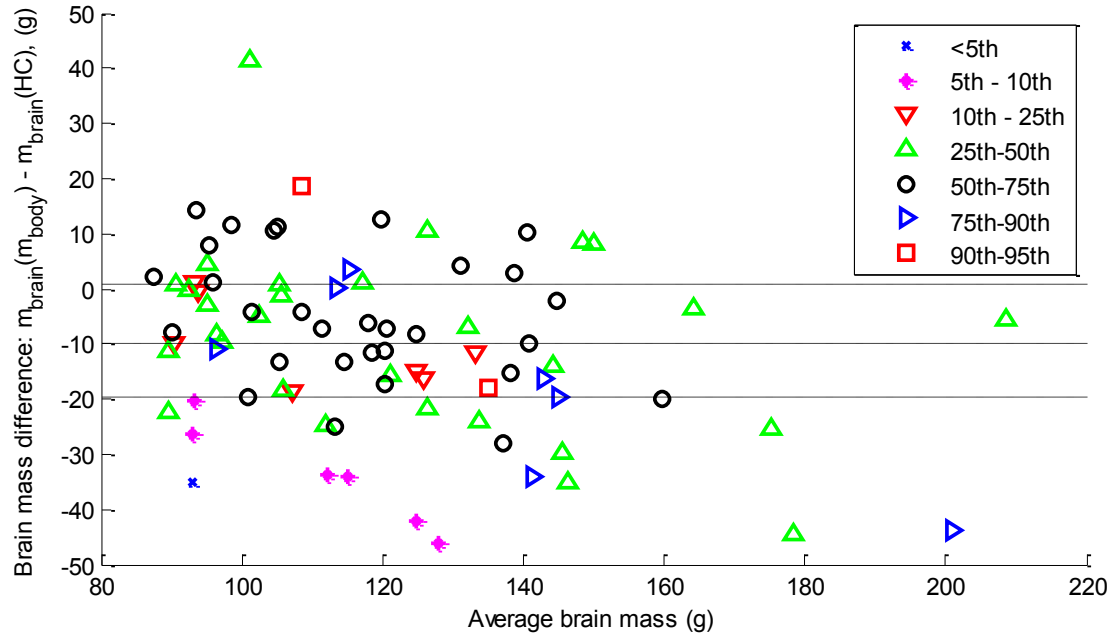


Figure 7.2: Comparison of BM and HC model estimations for brain mass at different birth weight percentiles

For the SGA group, the BM model fit SI was 2.9% lower than the HC model fit SI, suggesting that model selection for brain mass estimation has greater impact in infants at the lower extremity of birth mass outcomes. This difference is not significant ($p=0.57$), but may still yield significantly different insulin interventions in model-based control.

SI is significantly higher in the SGA cohort, irrespective of the brain mass estimation model used ($p<0.001$). However, variability is a much more important factor in glycaemic control and forecasting than absolute SI (Chase et al., 2011b, Le Compte et al., 2010b), particularly for safety from hypoglycaemia. The IQR for SI in SGA infants is wider, suggesting that they are more variable and thus more difficult to control.

Table 7.6: Deviations in SI profiles under different models

	Entire cohort	SGA only	P-value
S_I with m_{body} model (Med [IQR]) (mL/mU/min)	14.1 [6.7 – 23.9] e^{-4}	18.7 [4.8 – 34.2] e^{-4}	$P<0.001$
S_I with HC model (Med [IQR]) (mL/mU/min)	14.2 [6.8 – 24.1] e^{-4}	19.3 [5.1 – 35.1] e^{-4}	$P<0.001$
p-value	0.47	0.57	
% ΔS_I (Med)	1.1 [0.5 – 1.8]	2.9 [2.7 – 4.0]	

7.3.3 Effect on Control

Results for virtual trials across the entire cohort are shown in Table 7.7. BG outcomes are not significantly altered between control situations A-D, with no difference in safety from hypoglycaemia (%BG<2.6) and negligible difference in performance assessed by the % time in band (% BG within the 4.0 -8.0 mmol/L band).

While median insulin rates remained similar between simulations, the IQR differs. Assuming that the HC model provides a more accurate indication of brain mass than the BM model, using the HC model in control results in smaller changes in insulin infusion rate (compare A to B in Table 7.7). Overall, control is slightly better if the BM model is used in control (compare A-B and C-D in Table 7.7). However, these improvements are not likely to be clinically observable.

Control is also assessed for the SGA cohort only, shown in Table 7.8. Time in band is not improved by using the HC model to estimate brain mass. This simulation of 7 patient episodes included 166 BG measurements over 622h (9% of total cohort hours). Time in band for this group is significantly lower across all simulations, indicating another underlying reason for difficulty in control. It is worth noting that SGA infant data ($N = 7$), contributing only 9% of total hours, is reasonably sparse and thus more greatly influenced by outlying patients, rather than indicating a lesser performance.

Table 7.7: Control interventions and outcomes from virtual trials for the entire cohort.

Whole cohort statistics	A. Simulate HC, Control HC	B. Simulate HC, Control m_{body}	C. Simulate m_{body} , Control HC	D. Simulate m_{body} , Control m_{body}
Median insulin rate [IQR] (U/kg/hr):	0.04 [0.03–0.06]	0.04 [0.02– 0.07]	0.04 [0.02– 0.07]	0.04 [0.03–0.07]
% BG within 4.0–8.0 mmol/L	62.1	62.4	62.2	62.5
% BG > 10 mmol/L	13.3	13.2	13.3	13.2
% BG < 4.0 mmol/L	1.1	1.1	1.1	1.1
% BG < 2.6 mmol/L	0.1	0.1	0.1	0.1

Table 7.8: Control interventions and outcomes from virtual trials for the SGA cohort

Whole cohort statistics	A. Simulate HC, Control HC	B. Simulate HC, Control m_{body}	C. Simulate m_{body} , Control HC	D. Simulate m_{body} , Control m_{body}
Median insulin rate [IQR] (U/kg/hr):	0.05 [0.00–0.10]	0.05 [0.00–0.10]	0.05 [0.00–0.10]	0.05 [0.00–0.09]
% BG within 4.0–8.0 mmol/L	42.9	42.9	43.1	45.6
% BG > 10 mmol/L	28.9	28.9	30.0	28.9
% BG < 4.0 mmol/L	2.2	2.2	2.2	1.9
% BG < 2.6 mmol/L	0.3	0.3	0.3	0.3

The matched difference in insulin interventions between Cases A-B and A-D are shown in Figure 7.3 and summarised in Table 7.9. Comparing $A-B$ shows the difference between doses for either controller method, assuming that the HC model is true in simulation, while using HC (Case A) or m_{body} (Case B) in model-based control. Comparing $A-D$ shows the difference between best-case model selections, with no mismatch between virtual patient and controller model. For both comparisons, Case A is most likely to give equal or slightly more insulin (94.6 – 100% across comparisons and sub cohorts).

Table 7.9: Comparison of insulin interventions in simulation

Case comparison	Cohort	Percentage of interventions		
		Lower than A	Equal	Higher than A
A – B	AGA	0.3	99.4	0.3
	SGA	0.0	100.0	0.0
A - D	AGA	1.7	95.8	2.5
	SGA	4.0	89.6	6.4

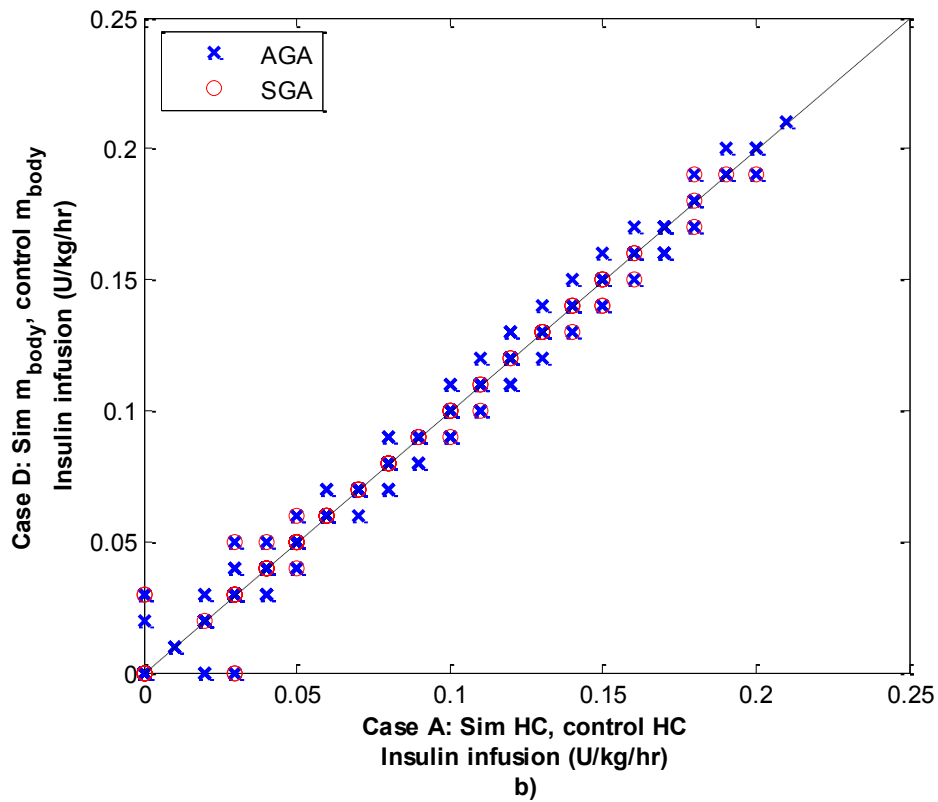
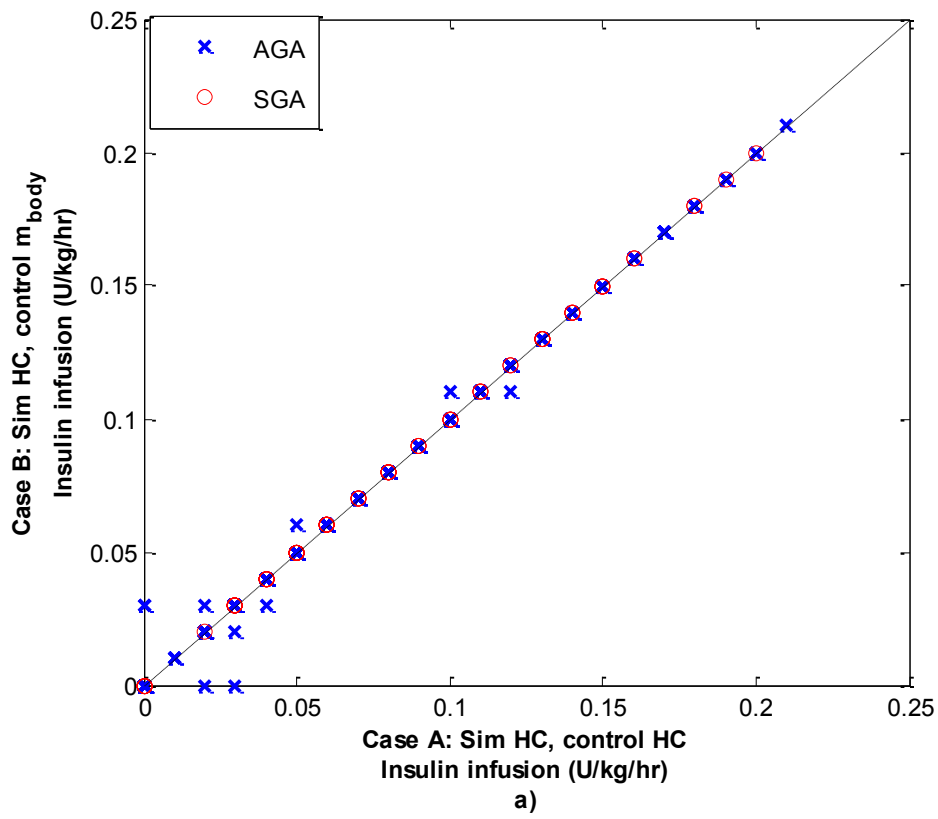


Figure 7.3: Matched insulin interventions by birth weight cohort for a) cases A vs. B, and b) cases A vs. D

7.4 Discussion

The HC model resulted in higher estimations of brain mass than a simple body mass based model. This effect was seen more clearly in infants that were SGA. Across the entire cohort, parameter changes in SI were clinically negligible. Control performance remained relatively unchanged or slightly worse when using the HC model for control decisions. This result was independent of which model was used in simulation of the virtual patient. Using the HC model resulted in more insulin being recommended for the same glycaemic outcomes, indicating that insulin was used less effectively.

The introduction of the Cooke method using HC would require an additional input for clinical staff when starting glycaemic control. Adding complexity to clinical protocols may increase clinical burden and potential for error (Aragon, 2006, Chase et al., 2008b). As a result, this analysis indicates that there is no control or clinical benefits to be gained from using the HC model instead of a body mass based model in this glycaemic control system.

Virtual trial performance is slightly worse than seen in Chapter 6. This result is likely because the data acquired from the HINT trial has some longer periods between interventions than those for which NICING was designed. In addition, the treatment of daily nutrition totals as constant parenteral or enteral infusions also degrades data quality. These differences can increase identified SI variability compared to the unknown reality and make control in virtual trials harder than would be seen clinically. However, in each case, these virtual trials are comparing the patients and cohort as their own control, thus eliminating this factor.

As the comparative accuracy of both the HC and body mass models is not known it would be beneficial to have an objective study compare the two models against a more accurate method. Head circumference data has not yet been analyzed for patients undergoing STAR at Christchurch Women's Hospital NICU, or any other cohort with more frequent BG measurement, which eliminated other sources of clinical data in this analysis.

The discrepancies observed between SGA and AGA infants are of interest. However, more data on SGA infants is necessary to claim robust conclusions. In particular, this investigation was not able to distinguish differences in symmetric and asymmetric SGA infants due to lack of data. As SGA infants are the most affected by model choice, this distinction may be of clinical use. Particularly as SGA infants were more insulin sensitive and variable in this small

cohort analysis. This difference, if verified, could increase risk in dosing. Future research should investigate these cohorts in greater detail.

Overall, the HC model is much more likely to be physiologically accurate. This outcome is due to the nature of the measurement and because the method is derived from a cohort of the same demographics it will be used on. However, metabolically its impact may not necessarily affect clinical outcomes in model-based insulin control.

7.5 Summary

The HC model resulted in higher estimations of brain mass than a simple body mass based model. This effect was seen more clearly in infants that were SGA. However, model based SI was not significantly different when fitted when the different brain mass estimation models in both SGA and AGA cohorts. A control based analysis showed that body mass based estimation of brain mass performed better. Thus, there is no control or clinical benefits to be gained from using the head circumference model instead of a body mass based model in this glycaemic control system. The SGA cohort has higher and more variable model-based SI, so potential differences in brain mass proportions between SGA and AGA infants are of interest to improve control. While this analysis showed no benefit in the use of head circumference based estimates, more data is needed for more comprehensive analysis.

7.6 Acknowledgements

The author wishes to thank C.A Gunn, of the AIC10 final year project team, who carried out the initial brain-mass analysis using a previous iteration of the NICING model under my supervision and guidance.

Chapter 8: Modelling Intestinal Glucose Absorption

8.1 Introduction

Postnatal growth restriction is a problem in premature infants, and in extremely low birth weight (ELBW) infants in particular (Clark et al., 2003b). The American Academy of Paediatrics suggests that nutrition goals should aim to achieve a postnatal growth rate approximating that of a normal foetus of the same GA (American Academy of Pediatrics Committee on Nutrition, 1985, Cooke, 2003). This nutrition can be delivered via parenteral or enteral routes, and there is significant debate in literature around appropriate onset and clinical procedure for enteral feed initiation (Karagol et al., 2013).

Enteral feeding of milk causes changes in gut structure and function, even in very premature infants (Aynsley-Green, 1983). These changes in gut structure and function include changes in gut motility, enzyme secretion, absorptive capacity, and endocrine response to nutrient absorption (Aynsley-Green, 1983). Early enteral feeding has been shown to decrease gut atrophy (breakdown of tissues) and increase intestinal permeability (Rouwet et al., 2002), as well as decreasing the incidence of sepsis (Flidel-Rimon et al., 2004).

In contrast to these positives, historically, there have been concerns about early enteral feeding causing necrotising enterocolitis (NEC) (Cakmak Celik et al., 2007, Ramani and Ambalavanan, 2013). However, many studies and randomised control trials have demonstrated no increase in the risk of NEC with early enteral feeding (Cakmak Celik et al., 2007, Caple et al., 2004, Hamilton et al., 2014, Karagol et al., 2013, Krishnamurthy et al., 2010, Rayyis et al., 1999).

Therefore, enteral feeding is very common in many neonatal intensive care units (Cormack et al., 2013, Patole and Muller, 2004, Ramani and Ambalavanan, 2013). As such, a model of glucose appearance from enteral feeds is necessary for tight glycaemic control. This chapter uses retrospective continuous glucose monitor (CGM) data in term and premature infants to model the appearance of glucose in the blood from enteral nutrition. In particular, the half-life of gastric absorption of glucose from the gut into the bloodstream is determined to better model glucose metabolism in the preterm neonate, and thus improve glycaemic control.

8.2 Methods

CGM traces reflecting glucose appearance from enteral nutrition administration were used to estimate gut absorption of glucose in premature and term infants. De-identified CGM data was available from premature and term infants in two clinical cohorts, one from a Neonatal Unit in Budapest, Hungary, and the second from a study carried out in New Zealand (Harris et al., 2013). Results were compared to estimates derived from reported gut absorption values in independent perfusion studies.

8.2.1 Adaptation of the NICING Glucose-Insulin Model for Older Neonates

The NICING model of glucose-insulin dynamics developed in Chapters 4 to 7 was used. This model has been developed for very premature infants, but is considered sufficiently reflective of premature and term infant physiology for use here in premature and term infants. The only points of change were that CNS uptake and fluid compartment values were allowed to change with GA and PNA, according to the literature-based models shown in Figures 8.1 and 8.2. Endogenous glucose production (EGP) was explored based on an expansion of the literature search of Chapter 6 for very preterm infants, but the median EGP was not found to differ consistently with GA, so EGP was maintained at the constant value of $EGP = 6.0$ mg/kg/.min.

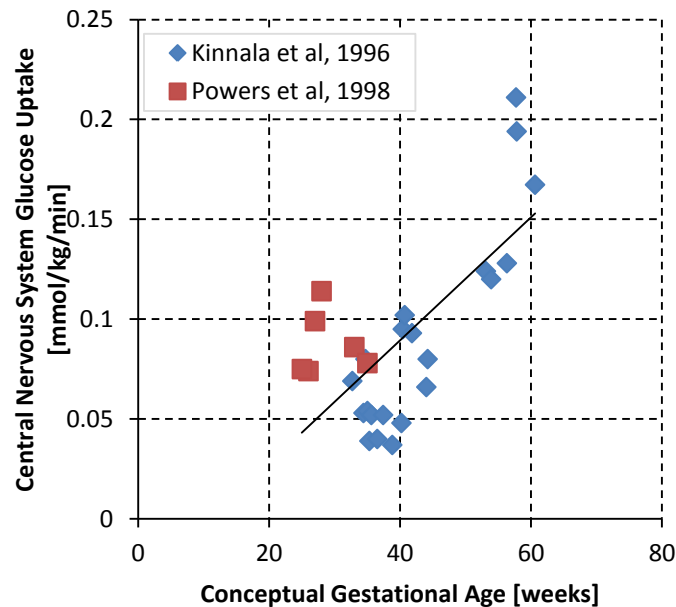


Figure 8.1: Increasing CNS uptake with increasing conceptual gestational age (CGA), where $CGA = GA + PNA/7$. Data sources: (Kinnala et al., 1996, Powers et al., 1998).

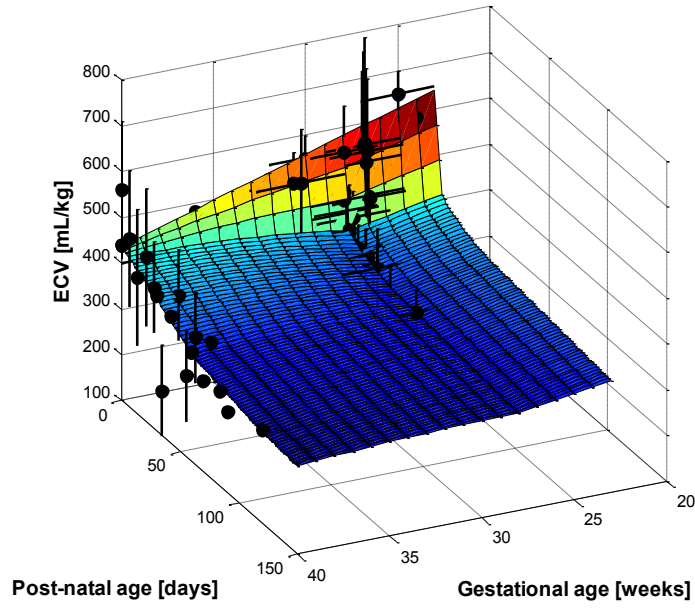


Figure 8.2: Extracellular fluid volume (ECV) with gestational and postnatal age. A drop in ECV in the first week following birth is observed across a wide range of studies, and is most prominent in the more premature infants. An ECV volume model is generated using a linear model derived from Avery et al. (1994) to give $ECV(PNA=0)$. $ECV(PNA>0)$ is described by a proportional drop off based on two long term studies of ECV in very preterm (~ 28 weeks) (Shaffer et al., 1986) and term (~ 40 weeks) (Friis-Hansen, 1954) infants. All data sources: (Coran et al., 1984, Friis-Hansen, 1954, Modi et al., 2000b, Shaffer et al., 1986, Shaffer and Meade, 1989a)

8.2.2 The Gut Model

The gut model comprises two compartments, P_1 and P_2 , denoting glucose in the stomach and intestine, respectively, which is a typical approach for these dynamics. The amount [mmol] of glucose in the stomach, P_1 , is defined:

$$\dot{P}_1 = P_{ex} - d_1 P_1 \quad (8.1)$$

Glucose is delivered to the stomach as glucose or glucose based sugar, and is denoted P_{ex} . Lactose is made up of glucose and galactose in roughly equal molar weight, and since galactose does not contribute directly to BG levels it was ignored. A value of $d_1 = 0.035 \text{ min}^{-1}$, corresponding to a half life of 20 min is the rate of gastric emptying of glucose from the stomach into the gut (Le Compte et al., 2010a). The amount of glucose in the gut, P_2 [mmol], is defined:

$$\dot{P}_2 = d_1 P_1 - \min(P_{max}, d_2 P_2) \quad (8.2)$$

Where d_2 [1/min] is the rate of uptake from the gut into the blood, and $P_{max} = 6.0$ mmol/L/min is an upper limit on this rate in adults (Lin et al., 2011).

8.2.3 Parameter Identification

The rate constant of glucose uptake from the gut into the bloodstream, d_2 , was the parameter of interest. SI and glucose appearance from the gut trade off mathematically, so the fitting process was iterative. The parameter d_1 was left constant as it also trades off with d_2 , and $d_1 = 0.035 \text{ min}^{-1}$ (corresponding to a 20 minute half life) is a reasonably acceptable value.

The NICING and gut models were initially fit using the adult value of $d_2 = 0.006 \text{ min}^{-1}$ (Lin et al., 2011) to get a S_I trace. The optimal d_2 values for each feed were then found using a grid search to minimise RMS fitting error over the range $d_2 = 0.001:0.060 \text{ min}^{-1}$. This process was iterated to update the S_I trace using the median d_2 value from the previous iteration. Iterations were continued until d_2 changed by less than 0.001 (3 decimal places, 2-3 iterations).

Several factors made parameter identification difficult. First, enteral feeds were recorded as having been given ‘on the hour,’ whereas in clinical practice the actual time of feeds is somewhere near this mark. For this reason, delivery time of the feed was varied by up to 20 minutes either side of the hour, and the d_2 with the lowest fitting error to the associated peak in the CGM data was selected. Second, feed duration was uncertain, and could vary between 5 and 20 minutes. While, for some patients, a reasonable estimate of feed time could be made based on delivery method, in other cases this estimate was not possible. Across the entire cohort the delivery duration was assumed a constant 12 minutes, and sensitivity of d_2 to delivery time was evaluated separately.

The parameter d_2 was analysed with respect to PNA, GA, BW, and total glucose content of feed to differentiate potential sources of variability between gut absorption in infants. Statistical comparisons are made using the non-parametric Mann-Whitney U test (Matlab[®], ranksum). P-values at $p < 0.05$ were considered significant.

8.2.4 Feed Methods

For the purpose of this investigation, enteral feeds were defined as any manually delivered (syringe/feeding tube) enteral nutritional intake delivered with duration of ~20min or less, involving expressed breast milk (EBM), nutritionally fortified EBM, or milk formula.

Breastfeeds were not considered due to uncertainty around the total fluid volume delivered, and rate of delivery. Enteral feeds delivered within 2 hours of a breastfeed, a change in parenteral (IV) nutrition, or oral dextrose gel use were discounted, to avoid any overlapping glucose appearance dynamics.

8.2.5 Clinical Data Cohorts

Budapest

CGM (Guardian REAL-Time, Medtronic, Minimed, Northridge, California) traces from 4 infants receiving 3 hourly enteral breast milk feeds of 20 mL or more were available from Semmelweis University, Department of Paediatrics, Hungary. Demographic data are in Table 8.1. According to the local ethical codes this analysis of retrospective CGM data is considered as a clinical data audit and the only ethics committee requirement was the depersonalization of the data collected.

SUGAR-BABIES

CGM traces (CGMS system gold, Medtronic, Minimed, Northridge, California) from 10 Patients from a cohort of 237 infants who were part of the SUGAR-BABIES study (Harris et al., 2013) were analysed. The SUGAR-BABIES study was approved by the Northern Y Ethics Committee, New Zealand (Harris et al., 2013).

The selection criteria included patients that were given 5 or more feeds of volume 20 mL or more at least 2 hours apart, with a clear rise and peak in the CGM trace corresponding to the timing of an enteral feed. The patient cohort is summarised in Table 8.1. Feeds were delivered over 5-20 minutes, depending on the feed method, but administration time was not recorded.

8.2.6 CGM Recalibration

CGM data gives 5 minutely readings of interstitial glucose concentrations, which are assumed to be in equilibrium with BG concentrations. CGM data for the SUGAR-BABIES study data was recalibrated to blood gas measurements of BG concentration using the measured current (ISIG) values and method in Signal et al. (2012). Recalibration was not carried out for the Budapest data, as not all necessary data was available.

Table 8.1: SUGAR-BABIES and Budapest patient cohort characteristics.

Patient	Gestational Age [weeks]	Birth Weight [g]	Hours after birth received first feed	Time after birth CGM was inserted	Feed type administered (for samples fitted)
<i>Budapest</i>					
1	36	3240	N/A	26 days	Breast Milk
2	40	4150	N/A	55 days	Breast Milk
3	39	3920	N/A	15 days	Breast Milk
4	40	5150	N/A	17 days	Breast Milk
<i>SUGAR-BABIES</i>					
1	39	3220	1.3	16.2 hrs	Term formula
2	36	2050	1.7	2.0 hrs	Preterm formula
3	38	2370	1.5	6.8 hrs	Mostly Term formula (2 mixed milk and formula)
4	36	2810	1.3	<1 hrs	Term formula
5	35	2260	0.7	<1 hrs	Mostly Term formula (1 mixed milk and formula)
6	31	1306	21.3	3.7 hrs	Breast milk
7	36	2140	4.4	1.7 hrs	Term formula
8	36	2135	3.5	6.5 hrs	Term formula
9	35	2450	0.5	<1 hrs	Term formula
10	33	2770	27.3	5.7 hrs	Breast milk

8.2.7 Reported Absorption Rates for Comparison

Glucose absorption rates for short intestinal sections were found in literature. These absorption rates were transformed into 'whole compartment' glucose absorption rates by assuming a linear glucose gradient down the gut, and integrating to give total glucose present in the gut [mmol]. This assumed model is shown schematically in Figure 8.3.

Assuming the glucose concentration profile is roughly linear (Borgstrom et al., 1957) in the gut

$$\check{P}_2 = \check{P}_{2s} - m x \quad (8.3)$$

Where \check{P}_2 is the local glucose concentration, \check{P}_{2s} is the glucose concentration at the start of the gut, x is position along the gut, and m is a constant.

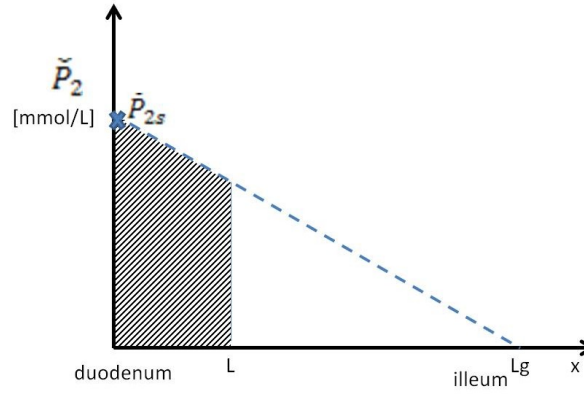


Figure 8.3: Linear glucose concentration across the small intestine.

Glucose is absorbed over a approximate length of $L_g = 3\text{m}$ (Borgstrom et al., 1957), and intestinal radius is $r=0.013\text{m}$ for adults (Helander and Fandriks, 2014). Small intestinal length in premature infants gives $L_g=0.145\text{m}$ (Touloukian and Smith, 1983) and $r=0.007\text{m}$ for infants (O'Neill, 2003). If at the L_g end the glucose concentration is $P_2 \approx 0$, then m is defined:

$$m = \frac{\check{P}_{2s}}{L_g} \quad (8.4)$$

The total absorption of glucose from the gut [mmol/min] is defined:

$$\int_0^{L_g} R_{ab} = d_2 c P_2 \quad (8.5)$$

Where P_2 is the total glucose in the gut, and is equal to the integral of the glucose concentration across the gut. The parameter c is the cross sectional area of the gut ($c=\pi r^2$). Thus, the total absorption across a given gut length, L_s , of the perfusion study is defined:

$$\int_0^{L_s} R_{ab} = d_2 c \int_0^{L_s} \check{P}_2 dx \quad (8.6)$$

Equation 8.6 can be integrated to give:

$$\int_0^{L_s} R_{ab} = d_2 c \left[\check{P}_{2s} L_s - m \frac{L_s^2}{2} \right] \quad (8.7)$$

To find d_2 , Equation 8.7 can be rearranged:

$$d_2 = \frac{\int_0^{L_s} R_{ab}}{c \left[\check{P}_{2s} L_s - m \frac{L_s^2}{2} \right]} \quad (8.8)$$

Equation 8.8 allows estimation of d_2 from intestinal glucose absorption studies in literature.

8.3 Results

8.3.1 Intestinal Glucose Absorption Constants from CGM Data

The calculated glucose absorption rate constant for all CGM data cohorts was $d_2 = 0.014$ [0.008 - 0.022] min^{-1} , corresponding to an absorption half life 50 [31 -86] min. Of the Budapest cohort, a d_2 solution was converged to in 77 of 165 sets of feed data. The median rate constant for glucose uptake was 0.014 [0.008 - 0.018] min^{-1} , corresponding to an absorption half life of 49 [39 - 86] min. Of the SUGAR-BABIES cohort, 178 sets of feed data across 10 patients converged to a solution, with a median [IQR] rate constant for glucose uptake of $d_2 = 0.018$ [0.012 - 0.026] min^{-1} , which corresponds to an absorption half life of 39 [27 - 58] min. In the SUGAR-BABIES cohort a further 140 sets of feed data were not used due to small feed size, failure to converge to a solution, failure of the feed to appear in the CGM trace, or a delay in the rise of BG indicating a more than 30 minute discrepancy between the recorded feed time and the given feed time.

Median and IQR varied between patients (Figure 8.4) with high variability over all patient metrics and no clear trend in d_2 over PNA ($R^2 < 0.001$), BW ($R^2 = 0.03$) or feed size (linear $R^2 = 0.03$).

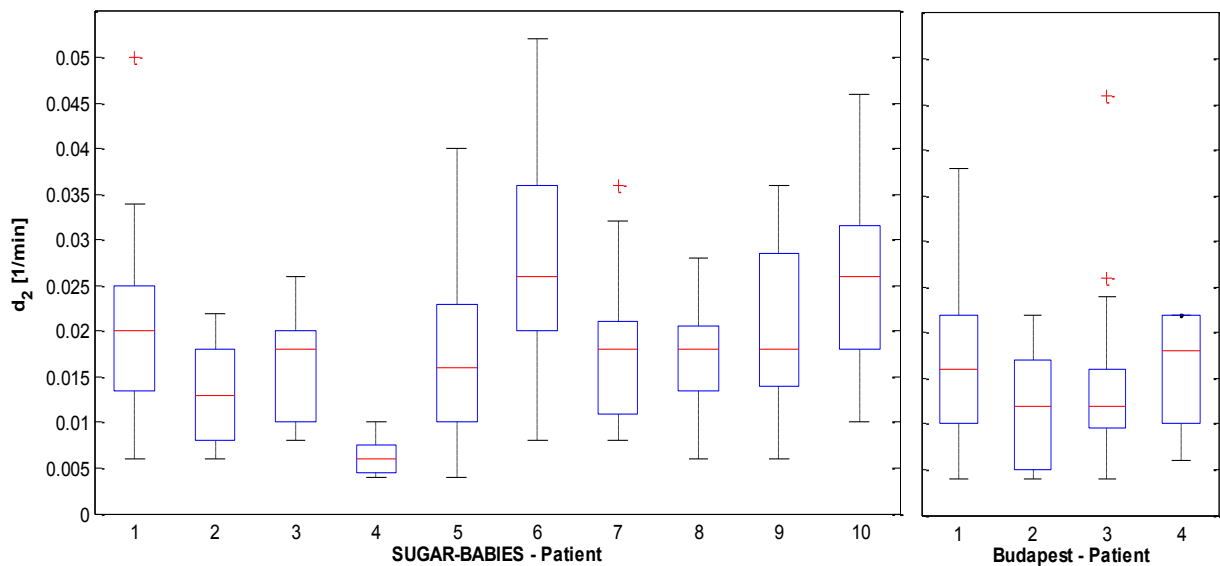


Figure 8.4: Per patient gut glucose absorption constants (d_2) across the SUGAR-BABIES and Budapest cohorts.

Table 8.2: Glucose absorption constants with gestational age (GA).

Grouping by Gestational Age: 36 week cut off (excluding breast milk patients)					
		Rate absorption constant, d_2 [min^{-1}]			
		GA < 36 weeks	GA = 36 weeks	GA > 36 weeks	GA \geq 36 weeks
N patients		2	4	2	6
N feeds		40	58	23	81
Median		0.017	0.015	0.018	0.018
[IQR]		[0.011 - 0.024]	[0.008 - 0.020]	[0.012 - 0.022]	[0.010 - 0.020]
p	GA < 36		<0.06	0.9	0.18
	GA = 36			0.15	0.28

In the SUGAR-BABIES data, for breast milk feeds (N=57), the median absorption rate constant was $d_2 = 0.026$ [0.018 - 0.035] min^{-1} (absorption half life of 27 [20 - 39] min) compared to the formula feeds (N = 118) value of $d_2 = 0.018$ [0.010 - 0.020] min^{-1} (absorption half life of 39 [34 - 69] min), ($p < 0.001$). The remaining 3 feeds were a mixture of breast milk and formula.

The gut absorption rate constant was, in general, higher with lower gestational age, but the relationship was weak ($R^2 = 0.05$). In the SUGAR-BABIES formula fed data (Table 8.2), using 36 weeks as the cut off age for three groups, because they approximate tertiles by GA and patient feed density, the gut absorption constant was not significantly different between infants grouped by GA ($p \geq 0.06$).

Figure 8.5 shows model fit for a representative selection of CGM trace sections. It can be seen that the model achieves a qualitatively good fit in most cases. However, it cannot completely capture the peak and/or trough of the glucose appearance. In addition, there may be un-modelled dynamics, unrecorded inputs, or sensor error in cases where multiple peaks are not captured.

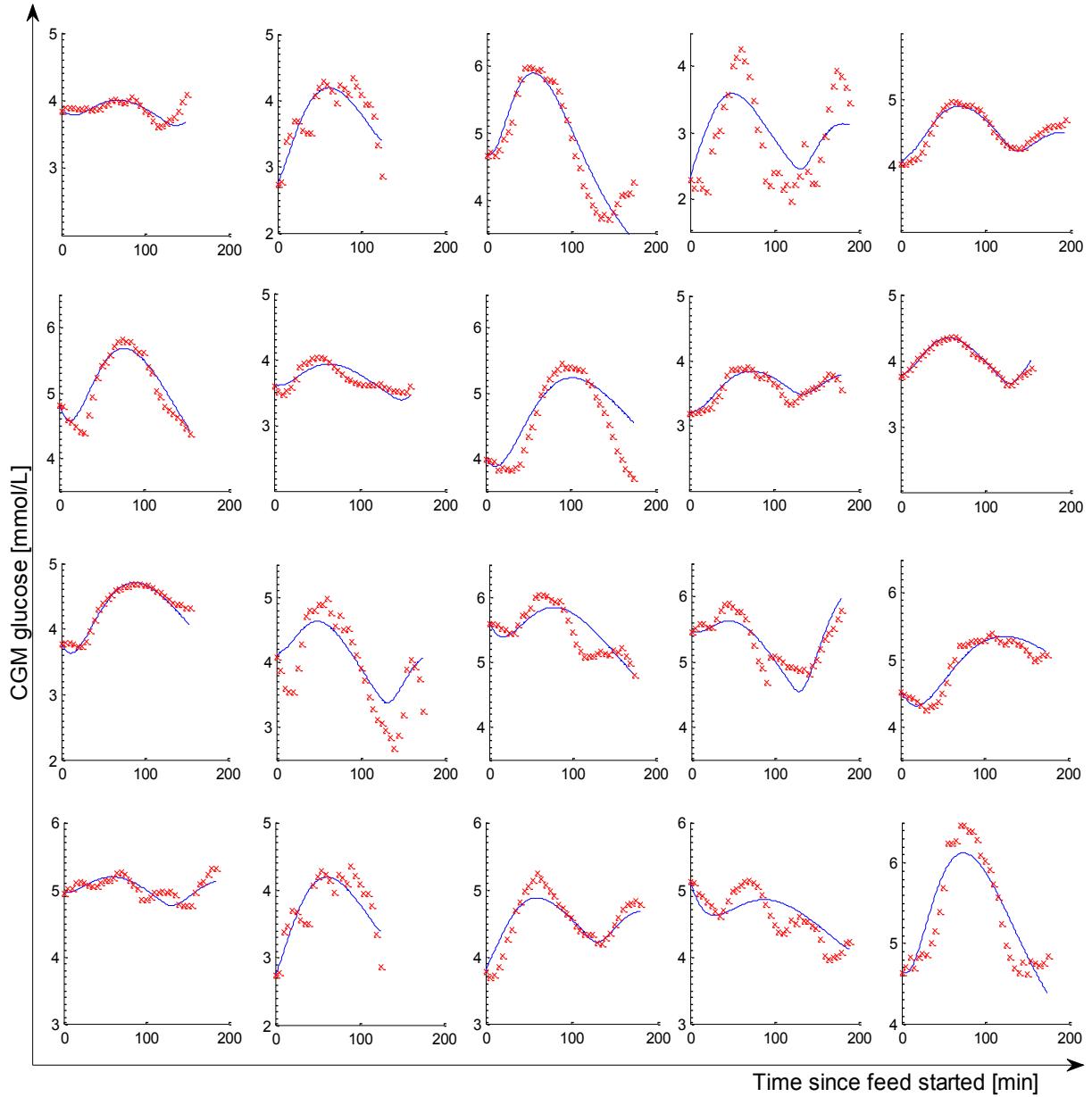


Figure 8.5: Fitting error over a representative selection of CGM trace sections from SUGAR-BABIES data. The crosses (x) show CGM data, and the solid line (-) the model solution around the time of an enteral feed.

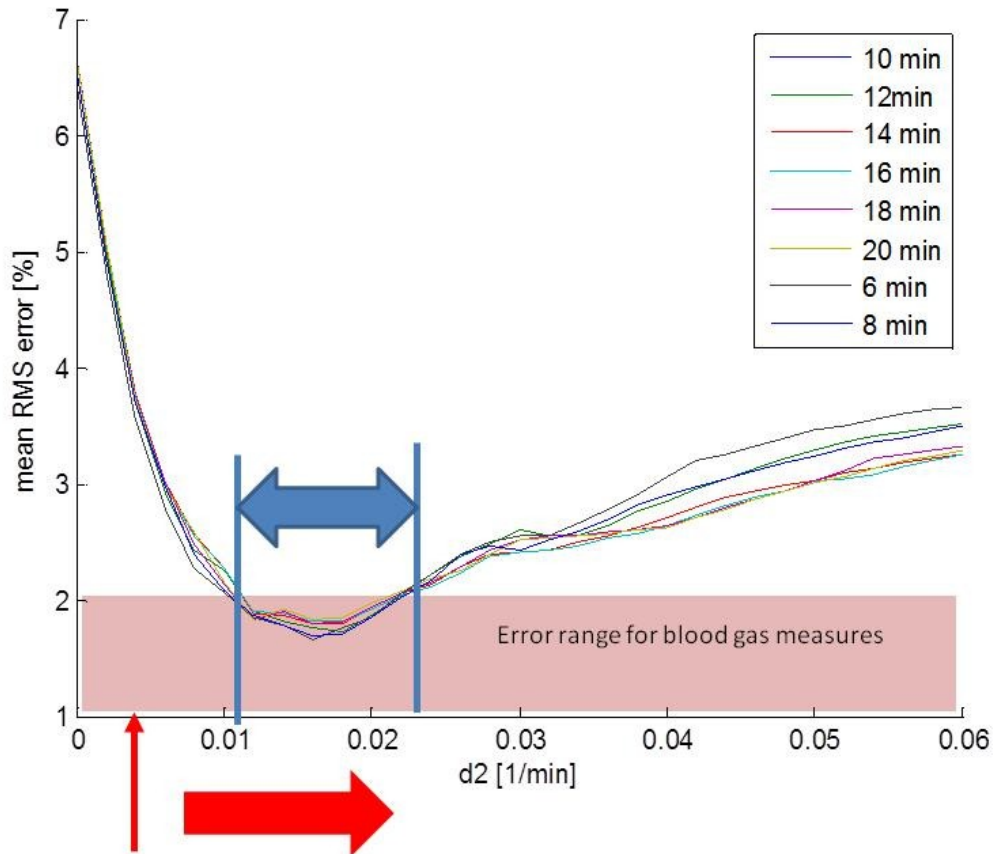


Figure 8.6: Fitting error across the d_2 range for different feed administration periods.

Sensitivity to the time over which the feed was administered was tested, and the results are shown in Figure 8.6. Fitting error does not differ significantly between different assumed administration times. Given that blood gas measures have an associated error of $\sim 2\%$, from Figure 8.6 the optimum d_2 can be estimated to lie between 0.011 and 0.024 min^{-1} , corresponding to a half life of 30 - 70 minutes.

The trade off between the gut model transport parameters d_1 and d_2 is shown in Figure 8.7. While there is a global minimum, the parameters trade off significantly within the middle of their likely physiological range. The global minimum exists in most cases at the upper end of the d_1 parameter range, suggesting short gastric emptying times, shorter than reported elsewhere in literature. For example, in Figure 8.7 the global minimum occurs at $d_1 = 0.040$, corresponding to a half life of 17 minutes. In contrast insulin sensitivity and d_2 trade off to a much smaller degree (Figure 8.8), with relatively small change in S_I for a relatively large change in d_2 .

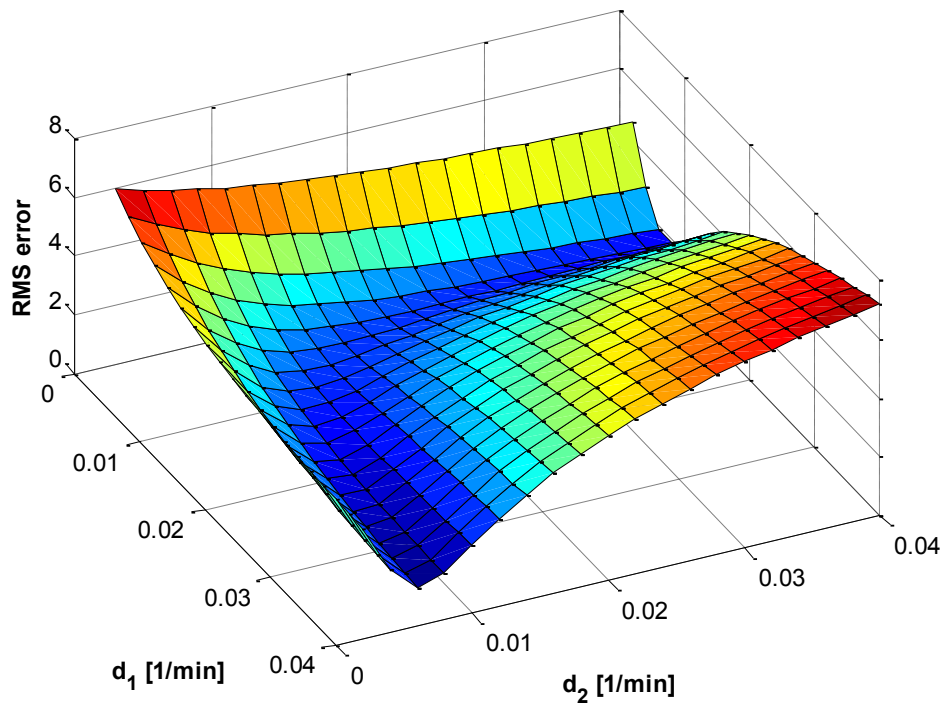


Figure 8.7: A typical error surface showing sensitivity and trade off of gastric emptying (d_1) and gut absorption (d_2) parameters for a two compartment gut model.

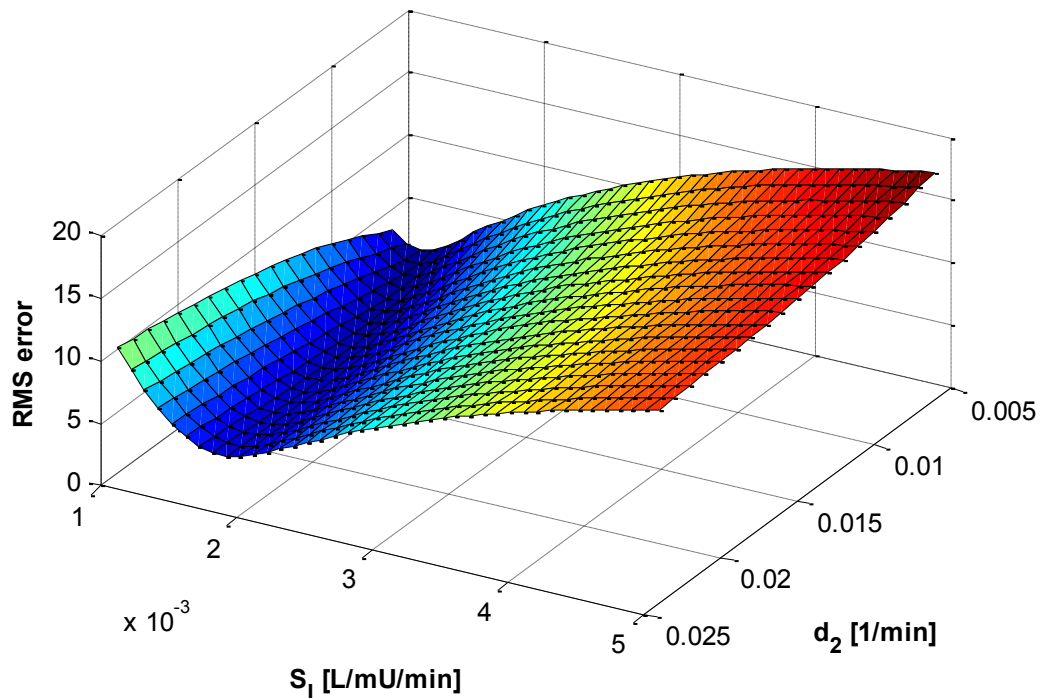


Figure 8.8: A typical error surface showing sensitivity and trade off gut absorption (d_2) and insulin sensitivity (S_1) parameters for a two compartment gut model.

8.3.2 Comparison to Reported Absorptions

Gut absorption rates were found in the literature for both adults (Modigliani and Bernier, 1971) and infants (Shulman, 1999) . These values were converted from a 'per study gut length' rate to a whole compartment rate constant, and the results are shown in Table 8.3. The average gut absorption rate constant is higher than the d_2 value determined here using CGM data. However, it is approximately the same order of magnitude.

Table 8.3: Intestinal glucose absorption rate constants derived from literature. Glucose infusion (P_{ex}) rate, initial glucose concentration (P_{2s}), and absorption rates (R_{ab}) found are used to derive a whole compartment absorption rate constant (d_2) using Equation 8.8.

	Study	P_{ex} [mmol/min]	P_{2s} [mmol/L]	$\int_0^{Ls} R_{ab}$ [mmol/min/25cm]	d_2 [1/min]
Adults	Modigliani and Bernier (1971)	0.5	66	0.472	0.056
		1.1	133	0.728	0.043
		2.1	260	0.82	0.025
		0.7	66	0.459	0.054
		1.3	133	0.763	0.045
		2.0	200	0.841	0.033
		2.6	260	1.019	0.030
		1.0	66	0.585	0.069
		2.0	133	0.885	0.052
		3.0	200	0.971	0.038
		3.9	260	0.999	0.030
		1.3	66	0.719	0.085
		2.7	133	0.913	0.053
		4.0	200	1.091	0.042
		5.2	260	1.042	0.031
	Median [IQR]	2.0 [1.2 - 2.8]	133 [100 - 230]	0.841 [0.723 - 0.985]	0.043 [0.032 - 0.054]
Preterm Infants (<37 weeks)	Study	P_{ex} [mmol/min]	P_{2s} [mmol/L]	$\int_0^{Ls} R_{ab}$ [mmol/min/15cm]	d_2 [1/min]
	Shulman (1999)	0.001	1	0.6	0.047
		0.01	10	3.6	0.028
		0.1	100	27.9	0.022
	Median	0.01	10	3.6	0.028

8.4 Discussion

8.4.1 CGM Derived Results

GCM data was used in a first ever analysis to estimate whole gut absorption of glucose in premature infants using a 2 compartment gut model. The median gut absorption rate constant was $d_2=0.014 \text{ min}^{-1}$, which corresponds to an absorption half life (time for half of total glucose to be absorbed) of 50 minutes. This value is twice as fast as the half life of 100 minutes used in the adult intensive care case. The glucose absorption rate constant was not found to differ significantly with GA. This result suggests that prematurity may not be a significant factor in glucose absorption, a result counter to expectations.

The median gut absorption constant here is of the same order as that calculated from adult and infant literature data (0.031 min^{-1} and 0.028 min^{-1} respectively, from Table 8.3), but slightly lower. This outcome seems to indicate the results derived from CGM data are physiologically likely, and that gut absorption in premature and term infants in intensive care may be lower than that in the literature for healthier infants due to their critically ill state, or feed composition differences (Bourlieu et al., 2014).

The practical insignificance of differences between absorption rate constants over GA and between the SUGAR-BABIES data and slightly older Budapest patients (Table 8.1) is further highlighted when blood gas error is considered. Figure 8.5 shows the RMS error is less than around 2% between $d_2 = 0.011$ and 0.023 min^{-1} (half life 30-63min), suggesting a wide valid range from which the glucose absorption can be selected. Almost all median values from Table 8.2 fall within this range, thus suggesting any observed differences between groups may result from chance measurement error.

Insulin sensitivity and glucose absorption in the gut trade off to a small degree. However, S_I is relatively insensitive to changes in the gut absorption constant d_2 (Figure 8.8). These results suggest that it is the total glucose absorbed rather than the rate that it is absorbed at that has the biggest influence on insulin sensitivity, and while a faster d_2 improves model fit to the more measurement dense CGM data, variability in d_2 does not significantly influence glycaemic control.

8.4.2 Contributions of Gastric Emptying and Feed Composition

A gastric emptying half life of 20 minutes ($d_1=0.035 \text{ min}^{-1}$) was used. This value is within the range of 17 – 60 minutes reported in the review by Bourlieu et al. (2014), where high variation in the rate of gastric emptying was seen within and between studies. If higher gastric emptying half lives of 30-40 minutes were used, fitting error increased (see Figure 8.7), as the width of the modelled glucose appearance exceeded that observed in the CGM data. This result suggests that, although d_1 and d_2 trade off to a large extent, there is a lower limit on d_1 . Gastric emptying is higher in premature infants than term infants Bourlieu et al. (2014), so while in this analysis a gastric emptying half life of 20 minutes was appropriate for term and premature infants, a longer half life of 30-45 minutes (Bourlieu et al., 2014) may be more appropriate in very and/or extremely premature infants.

For the SUGAR-BABIES cohort infants feed formula had higher glucose absorption half lives, which is a bias likely resulting, at least in part, from the assumption of a constant gastric emptying over the entire cohort. Gastric emptying is dependent on the enteral milk feed composition and caloric composition, and gastric emptying of human milk feeds is observed to be faster than that of formula milk feeds (Bourlieu et al., 2014). However, as fitting error increased with a higher gastric emptying time, it is also possible that milk composition can affect glucose absorption and/or the two compartment model is unable to adequately capture some of the GCM observed gastric dynamics.

In contrast, the glucose absorption rate constant for the breast milk fed Budapest cohort was lower than both the formula fed and breast milk fed cohorts. This constant could be lower due to the inability to recalibrate the CGM data, smaller cohort size (4 infants vs. 10 in the SUGAR-BABIES cohort), or cohort specific differences. However, the resultant median and inter-quartile range for this cohort falls within the 2% error region of Figure 8.5, indicating it may not be possible to separate these effects from sensor noise or blood gas measurements.

Given the wide range of possible glucose absorption rate constant values within the 2% error threshold, the study median of $d_2 = 0.014 \text{ 1/min}$ is a reasonable approximation of glucose absorption dynamics across a wide range of premature infants. This value is also a reasonable approximation of glucose absorption in very premature infants, in lieu of more gestational age specific data. However, since gastric emptying is known to be slower in very premature infants, the gastric emptying rate constant d_1 can be tailored to this cohort with a value of $d_1 = 0.017 \text{ min}^{-1}$, corresponding to a gastric emptying half life of 40 minutes.

8.4.3 Study Limitations

There are several limitations to this study. First, CGM data is well known for both sensor noise and drift (Castle and Ward, 2010, Facchinetti et al., 2014, Facchinetti et al., 2010, Kuure-Kinsey et al., 2006). For each feed, the initial glucose value used was the initial CGM BG value, rather than the model fit up to that point, which minimised the effect of sensor drift. In addition, CGM values for the SUGAR-BABIES cohort were recalibrated according to the method of Signal et al. (2012), which improves the correlation between sensor data and BG measures. Around 44% (140) of the total feeds in the data failed to converge to a solution or failed to display a clear peak in the CGM data around the time expected from the feed. As a result, the results in this paper should be considered indicative only of glucose absorption, with glucose absorption being variable over time and between patients, as might be expected in any case.

Additionally, infants were selected for this study primarily based on feed volume and number of feeds. Thus, infants that were likely to display the largest feed related spikes in CGM data were chosen, and so the gut absorption constants found may best reflect relatively large feeds and/or faster absorption, the latter of which could skew model-based results. Table 8.3 shows that the glucose uptake rate is not in direct proportion to the gut glucose concentration. The results in this paper display no trend and high variability with feed size and glucose concentration, and thus do not confirm, but also, do not rule out, different gut absorption constants that vary with different feed sizes.

A third potential limitation is the use of the NICING model developed for very low birth weight premature infants, in premature and term infants. Fluid volumes and central nervous system uptake were tailored to the infants in this study based on their gestational and postnatal age. Without the relatively rare data to tailor insulin clearances and secretion to this older cohort, the NICING model was considered to be sufficiently reflective of neonatal physiology for the purposes of this study.

Finally, the CGM data in this study is from older infants than those the gut absorption model is being designed for. This difference is perhaps partly offset through the use of the NICING model. However, given the lack of such CGM data being available for very or extremely premature infants, the results are considered more indicative of premature infant gut glucose absorption than that of adult ICU patients. Thus, these results are sufficient for an estimation

of the gut glucose absorption rate constant, but should be interpreted with caution outside the context of this study and the NICING model.

8. 5. Summary

CGM data has been used to estimate the half life, and therefore the rate constant, of glucose absorption from the gut to the bloodstream. The median gut absorption rate constant was $d_2 = 0.014 \text{ min}^{-1}$, which corresponds to an absorption half life of 50 minutes. The glucose absorption constant was found to be higher in formula fed infants than breast milk fed infants in the SUGAR-BABIES cohort. However, this constant was lower in the breast milk fed Budapest cohort in comparison to both breast milk and formula fed SUGAR-BABIES cohorts. Given the range in $d_2 = 0.011 - 0.024 \text{ min}^{-1}$ within the 2% measurement error threshold, the median value of $d_2 = 0.014 \text{ min}^{-1}$ is a reasonable approximation for a whole-cohort value, given the limitations of the study.

The gastric emptying rate constant was set to $d_1 = 0.035 \text{ min}^{-1}$ (20 minutes half life) for this analysis in term and premature infants, which is at the lower end of the range reported in literature. Increasing the half life of gastric emptying increased fitting error in this analysis. Given that gastric emptying is slower in very low birth weight premature infants, it is recommended that the gastric emptying constant be set to $d_1 = 0.017 \text{ min}^{-1}$ (40 minutes half life). It could also be varied with formula type.

8. 6. Acknowledgements

Dr. Deborah Harris from the Waikato District Health Board and Dr Gábor Marics from Semmelweis University for providing the data.

Chapter 9: Model Summary

Chapters 3 to 8 have attempted to better capture inter- and intra- patient metabolic variability through better modelling of metabolic processes. This task was done using available data from the extremely fragile very premature infant cohort, and the focus has been on developing models to much better capture and describe glucose-insulin kinetics in a clinical setting using common bedside measures. This new model is known as the Neonatal Intensive Care Insulin-Glucose-Nutrition (NICING model).

The NICING model differs from its predecessor, developed by (Le Compte, 2009), in several important aspects:

- An insulin secretion model has been developed specifically for the very/extremely premature infant cohort (Chapter 4).
- Insulin clearance pathways have been differentiated, and model parameters are made specific to the very/extremely premature infant (Chapter 5).
- Differing compartment distribution volumes for insulin are now taken into account between the interstitial fluid space and blood plasma (Chapter 5).
- Endogenous glucose production was extensively investigated, but remains a population constant, as discussed in Chapter 6.
- The previous model of glucose absorption via the gut has been replaced with a new model that conserves mass. Model parameters based on premature/term infant data have been identified (Chapter 8).

This short chapter summarises the modal equations and parameters in one place.

9.1 Neonatal Intensive Care Insulin-Glucose-Nutrition (NICING) model

9.1.1 Insulin

Insulin kinetics are modelled via two compartments, plasma insulin (I) and interstitial insulin (Q), described:

$$\dot{I} = -\frac{n_L I(t)}{1 + \alpha_I I(t)} - n_K I(t) - n_I (I(t) - Q(t)) + \frac{u_{ex}(t)}{V_P m_{body}} + (1 - x_L) u_{en}(t) m_{body} \quad (9.1)$$

$$\dot{Q} = n_I \frac{V_P}{V_Q} (I(t) - Q(t)) - n_C \frac{Q(t)}{1 + \alpha_G Q(t)} \quad (9.2)$$

Insulin secretion is modelled as a piecewise linear function of blood glucose concentration (G) that differs with infant sex:

$$u_{en} = \begin{cases} \max(4.2, -1.5 + 1.9 * G) & \text{if female} \\ \max(2.2, -0.37 + 0.86 * G) & \text{if male} \end{cases} \quad (9.3)$$

Patient specific and/or time varying insulin clearance parameters are defined:

$$n_C = n_I \frac{V_P}{V_Q} \left(\frac{I_{ss}}{Q_{ss}} - 1 \right) \quad (9.4)$$

$$GFR = 0.45 + 0.24 m_{bw} + \frac{0.18 PNA}{7} \quad (9.5)$$

$$n_K = \frac{1}{V_P} \times \frac{0.9 GFR}{0.6} \quad (9.6)$$

PNA is postnatal age in days. Insulin distribution volumes in the plasma and interstitial compartments are approximated by plasma (V_P) and interstitial (V_Q) fluid volumes respectively. These volumes are estimated as:

$$V_P = 0.047 L/kg \quad (9.7)$$

$$V_Q = 492 \times PNA^{-0.09} - V_P \quad (9.8)$$

All parameter descriptions and values can be found in Table 9.1.

Table 9.1: Insulin kinetic parameters

Parameter	Corresponding physiology	Value
α_I	Saturation on liver clearance of insulin	0.0017 L/mU
n_L	Liver clearance of Insulin	0.39 min ⁻¹
x_L	First pass liver clearance of insulin	0.67
n_K	Kidney clearance of insulin	Variable – Eq. 9.6 [min ⁻¹]
n_I	Diffusion of insulin between plasma an interstitial spaces	0.025 min ⁻¹
n_C	Clearance and degradation of insulin from interstitial space	Variable – Eq. 9.4 [min ⁻¹]
$u_{en}(t)$	Endogenous insulin secretion by pancreas	Variable – Eq. 9.3 [mU/L/kg/min]
$u_{ex}(t)$	Exogenous insulin therapy	Variable input [mU /min]
V_P	Fluid volume of plasma	0.047 L/kg
V_Q	Fluid volume of interstitial space	Variable [L/kg]
α_G	Saturation on insulin receptor binding	0.0 L/mU
m_{body}	Body mass	Variable [kg]
PNA	Post natal age	Variable [days]

9.1.2 Glucose

Blood glucose concentration, G , is modelled:

$$\begin{aligned} \dot{G} = & -p_G G(t) - S_I G(t) \frac{Q(t)}{1 + \alpha_G Q(t)} + \frac{P_{TPN}(t) + P_{EN}(t)}{V_{g,frac}(t)m_{body}} \\ & + \frac{EGP m_{body} - CNS m_{brain}}{V_{g,frac}(t)m_{body}} \end{aligned} \quad (9.9)$$

where brain mass may be estimated as 14% of body mass. Further parameter descriptions and values can be found in Table 9.2. It should be noted that in the case of EGP and CNS parameter values are given in both mg/kg/min and mmol/kg/min. These values are consistent with each other (glucose has approx 180g/mol), and reflect differing clinical practice in measuring blood glucose in mmol/L and glucose delivery in mg/kg/min. As EGP and CNS are constants, there are no scaling or modelling issues associated with the conversion of literature/clinical data units of mg/kg/min to units of mmol/kg/min in Equation 9.9.

Table 9.2: Glucose kinetic parameters

Parameter	Corresponding physiology	Value
p_G	Non-insulin mediated glucose uptake	0.003 min ⁻¹
S_I	Whole body insulin sensitivity	Variable [L/mU/min]
α_G	Saturation on insulin receptor binding	0.0 L/mU
$P_{TPN}(t)$	Intravenous glucose delivery	Variable [mmol/min]
$P_{EN}(t)$	Glucose absorption from gut	Variable [mmol/min]
$V_{g,frac}(t)$	Distribution volume of glucose	Variable [L/kg]
EGP	Endogenous glucose production	0.033 mmol/kg/min (15.8 kg/kg/min)
CNS	Glucose uptake by central nervous system	0.088 mmol/kg/min (15.8 kg/kg/min)
m_{body}	Body mass	Variable [kg]
m_{brain}	Brain mass	Variable [kg]

9.1.3 Gut Model

Glucose in the stomach, P_1 , and gut, P_2 , from enteral feeds is modelled by a two compartment model:

$$\dot{P}_1 = -d_1 P_1 + P(t) \quad (9.10)$$

$$\dot{P}_2 = -\min(d_2 P_2, P_{max}) + d_1 P_1 \quad (9.11)$$

$$P_{EN}(t) = \min(d_2 P_2, P_{max}) \quad (9.12)$$

Parameter values and descriptions are given in Table 9.3. It should be noted that as the data for the analysis in Chapter 8 was made available after clinical implementation of the model described in 9.1.1 and 9.1.2. As a result, the value for the glucose absorption constant d_2 used in the clinical implementation was that used in the adult ICU ($d_2 = 0.007 \text{ min}^{-1}$). Further parameter descriptions and values can be found in Table 9.3.

Table 9.3: Glucose kinetic parameters

Parameter	Corresponding physiology	Value
d_1	Gastric emptying of glucose from stomach to gut	0.017 min ⁻¹
d_2	Glucose absorption from gut to blood stream	Old value: 0.007 min ⁻¹ New value: 0.014 min ⁻¹
P_{max}	Maximum glucose absorption rate	6.0 mmol/min
$P(t)$	Enteral feed rate	Variable [mmol/min]
$P_{EN}(t)$	Rate of glucose appearance in blood stream from gut	Variable [mmol/min]

9.2 Model Solution and Fitting

Model solutions are generated from Equations 9.1, 9.2, 9.9, 9.10 and 9.11 to give G , I , Q , P_I and P_2 using numerical integration and Matlab[®]ode45 numerical solver. Patient-specific time-varying insulin sensitivity, S_I , is fit on an hour to hour basis using integral based fitting (Hann et al., 2005). This process involves the integration and re-arrangement of Equation 9.9 to find S_I . If S_I is constant from hour t_i to t_{i+1} , then Equation 9.9 can be integrated and re-arranged to give:

$$S_I = \frac{-G|_{t_i}^{t_{i+1}} - \int_{t_i}^{t_{i+1}} p_G G(t) dt - \int_{t_i}^{t_{i+1}} \left(\frac{P_{TPN}(t) + P_{EN}(t)}{V_{g,frac}(t)m_{body}} + \frac{EGP m_{body} - CNS m_{brain}}{V_{g,frac}(t)m_{body}} \right) dt}{\int_{t_i}^{t_{i+1}} G(t) \frac{Q(t)}{1 + \alpha_G Q(t)} dt} \quad (9.13)$$

When fit over all hours in clinical data, a time-varying insulin sensitivity profile is generated. This forms the basis of a virtual patient in simulation, and is used to forecast future changes in insulin sensitivity in the STAR-framework (Section 3.4).

Linear interpolation of BG measures provides a first estimate of G in equation 9.13. The identification process can be made iterative, with the substitution of each new model generated G profile into 9.13 to re-identify S_I . However, it was found that S_I was not significantly different between iterations, and an interpolated G profile is sufficient for parameter identification of S_I .

9.3 Summary

This chapter briefly presents the NICING model in its final form, along with all parameter values and sub-models. This form of the NICING model, with a noted exception in terms of the new gut model, will be used in the next few chapters to design and test a control protocol in virtual trials, and implement it in a clinical context.

Chapter 10: Protocol Development and Optimisation

10.1 Introduction

Chapter 2 discussed different clinical approaches to insulin therapy. Surveys have shown that insulin therapy clinical practice differs significantly between different neonatal intensive care units, with differing thresholds for hyperglycaemia and insulin therapy onset (Alsweiler et al., 2007). Further, not all units have a protocol for dosing insulin (Alsweiler et al., 2007, Simeon and Gottesman, 1991). Insulin dosing protocols tend to be ad hoc, or insulin is titrated with BG level in a fixed or flexible protocol that, critically, ignores nutrition and insulin inputs. In addition, often a single glycaemia level is clinically targeted as the ideal, rather than a range of acceptable values, which doesn't account for intra-patient variability (Chase et al., 2011b).

Stochastic models were outlined in Chapter 3, based on previous work (Le Compte, 2009, Le Compte et al., 2010b, Lin, 2007, Lin et al., 2006). Given a current insulin sensitivity (S_I), a stochastic forecasting model then predicts the range of possible changes in SI over the coming intervention interval. From this range of SI values a range of resulting BG outcomes can be calculated for a given insulin intervention, as shown again in Figure 10.1. This calculation of a range of probable BG outcomes allows stochastic targeting protocols to utilise insulin therapy to actively target a desired target range or threshold with a specified level of hypoglycaemic, out of range, risk. The aim is to overlap these predicted BG outcomes with a clinically specified target range to maximise the likelihood of the BG levels in that range. This probabilistic target to range approach is unique in glycaemic control.

There are several ways by which the desired overlap of outcomes and the targeted BG range can be achieved. These approaches typically use various percentiles of likely outcomes and targeted range thresholds or values. This chapter will analyse several different stochastic targeting protocols, with the aim of developing an optimised protocol for effective and safe use in neonatal intensive care.

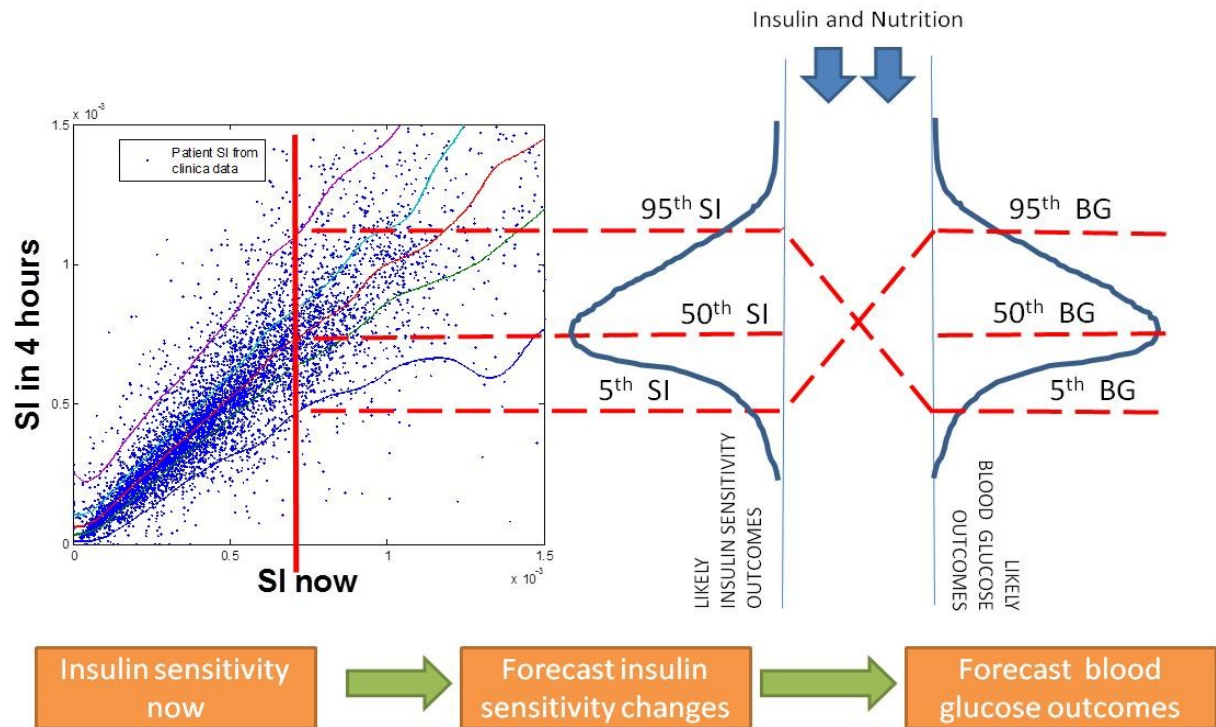


Figure 10.1: Stochastic forecasting of insulin sensitivity (SI) and blood glucose (BG) outcomes. First a patients current SI is determined. Distributions of future insulin sensitivity are generated from stochastic models, and these are used to generate distributions of BG outcomes for a given insulin/nutrition treatment (usually characterised by the 5th, 50th, and 95th percentiles).

10.2 Methods

10.2.1 Stochastic Targeted Protocol Designs

To determine a baseline SI, a 0.05 U/kg/hr insulin infusion is started and maintained for 2 hours. This is a relatively low and safe dose to provide a second BG measurement. BG is then measured, SI determined, and the STAR protocol calculates the next insulin intervention.

The previous STAR-NICU controller (Le Compte et al., 2009) checks insulin doses until the 50th percentile BG prediction is 6.0 mmol/L (Le Compte et al., 2011). It then checks that 95% of predicted BG levels $BG \geq 4.0$ mmol/L, and reduces insulin if required to meet this constraint, allowing median BG to rise. To improve this approach, the following control approaches were tested:

PROTOCOL 1: Select the maximum allowable insulin infusion rate that causes the 5th percentile BG prediction to be equal to or greater than a specified lower BG limit. This approach limits the risk of BG less than a lower target to 5%, but does not target the median result.

PROTOCOL 2: Select the maximum insulin infusion rate that places the 95th percentile BG prediction on or below an upper BG limit. This approach is more aggressive and does not consider a lower limit, except implicitly through choice of the upper limit.

PROTOCOL 3: As per Protocol 1, but giving an insulin bolus that yields a 5th percentile BG on or above a specified lower limit in the first hour, followed by an infusion for the remaining time period so that the 5th percentile BG remains on or above the specified lower limit at the end of the entire measurement interval.

PROTOCOL 4: As per Protocol 4, but giving an insulin infusion, instead of a bolus, over the first hour.

For each of the protocols a range of target values was tested. For Protocols 1, 3, and 4, the 5th percentile targets tested were 4.0, 4.2, 4.4, 4.8, and 5.0 mmol/L. For Protocol 2, the 95th percentile was tested on targets of 8.0, 9.0, 10.0, and 11.0 mmol/L. Protocols 3 and 4 allow more aggressive control where certainty is the greatest - in the first hour, immediately after SI is identified. Figure 10.2 systematically illustrates the four protocol approaches.

In all protocols, if measured BG < 4.0 mmol/L, no insulin is given to for safety. An upper limit is applied to the amount the insulin rate can increase between treatments to ensure control is not compromised by glucose meter error, or short perturbations from the neonate's normal glycaemic state. No attempt was made to regulate nutritional inputs, which were left to clinical choice, which is a major difference from the adult ICU.

Insulin infusions are increased in steps of 0.01 U/kg/hr, with a maximum allowable insulin infusion of 0.3 U/kg/hr, and a minimum pump rate of 0.1 mL/hr (~0.03 U/kg/hr). Insulin concentration was simulated according to the weight measurements in the patient data set, and the following clinical formula for concentration from Christchurch Women's hospital:

$$U_{conc}[U/mL] = \frac{5 * weight}{20} \quad (10.1)$$

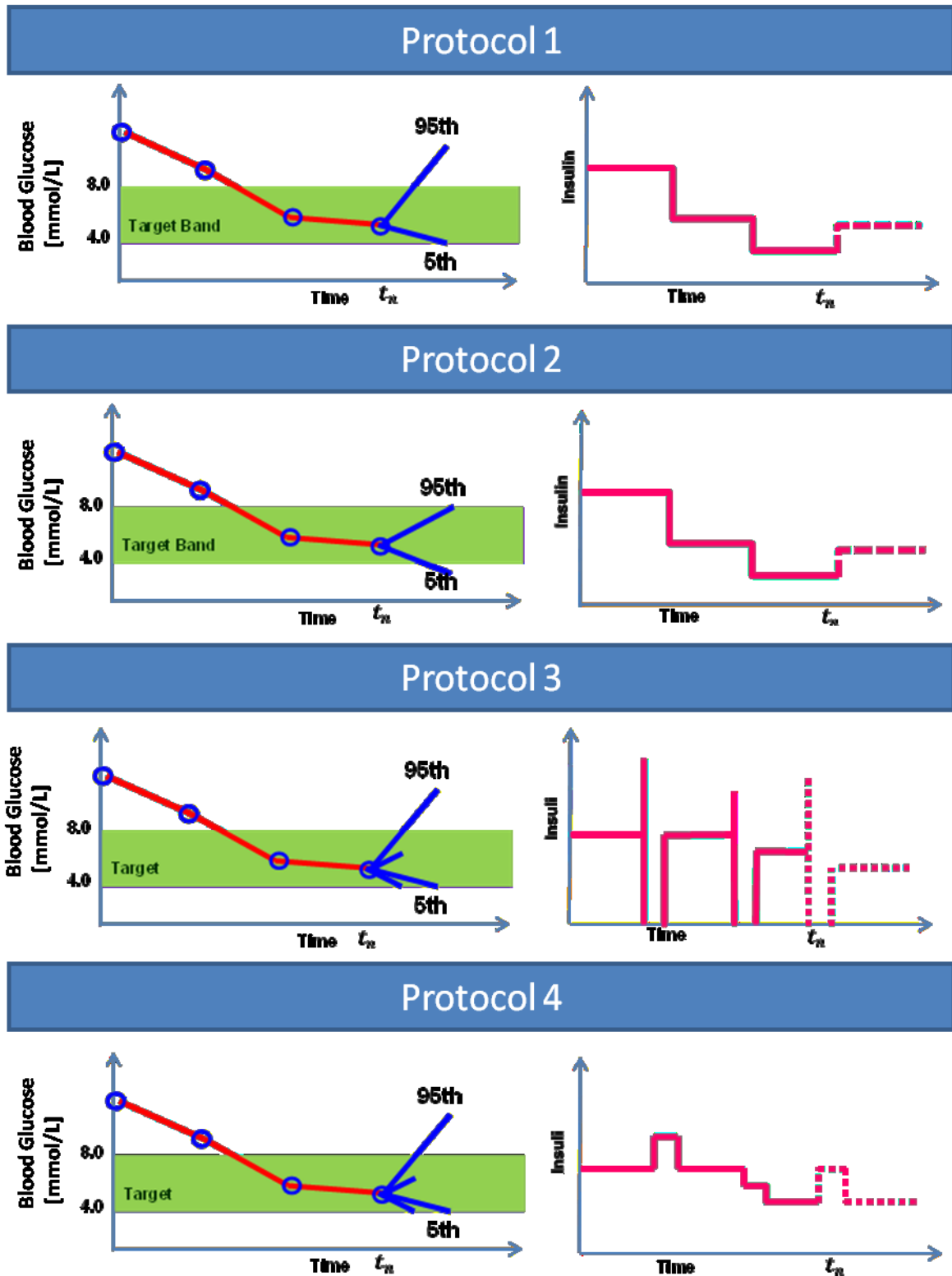


Figure 10.2: Stochastic targeting and treatment selection for Protocols 1-4. Dashed lines show the shape of the proposed insulin treatment from the current time, t_n . It should be noted that Protocols 3 and 4 examine stochastic bounds at 1 and 4 hours due to the initial treatment given.

To improve the best protocol, a sensitivity analysis was carried out on the limit placed on the increase in insulin rate between treatments, and a tolerance was applied to the targeted value. Limits on the rate of insulin change can limit control over-response to short-term changes in observed sensitivity to insulin caused by measurement device error or rapid fluctuations in patient condition. The limit used by the STAR-NICU controller is set to a maximum 0.03 U/kg/hr increase in insulin infusion. The effect of higher and lower limits was investigated, as well as variable, BG defined limits allowing greater changes at higher BG levels. These variable BG defined limits allowed a rate increase of 0.06 U/kg/hr for BG greater than a defined limit, maintained a limit on rate increase of 0.03 U/kg/hr within an intermediate range, and allowed a rate increase of 0.01 U/kg/hr below a defined BG limit, for safety reasons.

10.2.2 Virtual Trial Methods

The different protocols were tested using clinically validated virtual trial methods (Chase et al., 2010a). A virtual patient in this method consists of a SI profile fitted from clinical data using Integral-based fitting methods (Hann et al., 2005). As SI is treatment independent in the model ((Chase et al., 2010a) the known SI profile and NICING model can be used to describe what the BG response to insulin and nutrition would have been under different treatment doses. Model fit procedure is summarised in Section 9.2.

Real time control can be simulated in virtual trials using these virtual patients. The control protocol is made blind to the known SI profile after the simulated 'current time.' Using the SI at that point, stochastic forecasting can be used to predict a range of likely SI outcomes, which in turn guide the control protocol to select an insulin dose. Once the control protocol has made its treatment selection, the known SI profile can be used to determine the BG response to this treatment up until the next simulated BG measurement. Measurement noise, timing errors, and other variability may also be included in the simulation. The process is repeated until the end of a virtual patient episode is reached. This process is summarised in Figure 10.3.

10.2.3 Virtual Patient Data

The virtual patient cohort consisted of those patients described in Section 6.2.2, and their clinical characteristics can be found repeated in Table 10.1. Virtual patients were created from retrospective data, and clinical data from infants who were treated using the STAR-

NICU protocol. Retrospective data is comprised of 22 patients totalling 25 episodes, who received insulin under a bedside titration protocol prior to STAR implementation in 2008. STAR-NICU data was collected from August 2008 to December 2012, where 40 premature infants totalling 53 episodes received insulin under the STAR-NICU protocol, as either a pilot trial or as a standard of care, and 22 of these have been previously reported (Le Compte et al., 2012).

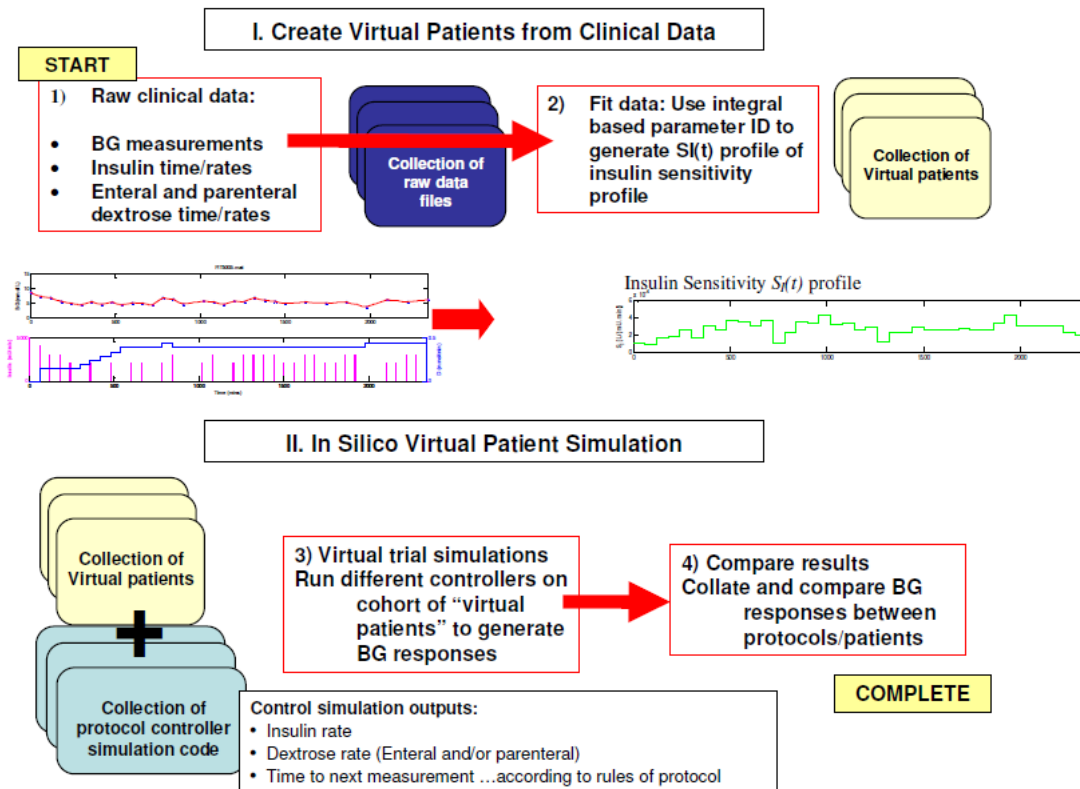


Figure 10.3: Virtual patient development and in silico simulation method. Virtual patients are created from clinical data (I), and then used in virtual trials (II) to test glycaemic outcomes of protocols. Figure sourced from (Chase et al., 2010a).

Table 10.1: Clinical characteristics of virtual patient cohort

Characteristic	Retrospective	STAR-NICU
n Patients (n male)	22 (7)	40 (22)
n Episodes	25	54
Total Hours	3098	5014
Number of BG measurements	943	1632
Gestational Age [weeks]	27 [25-27]	26 [25-29]
Weight [g]	845 [800-900]	785 [635-950]
Post Natal Age [days]	3 [1-7]	3 [1-12]

10.2.4 Analyses and Performance Metrics

Results were compared against the existing protocol. The following performance metrics were used:

- **SAFETY:** number of severe hypoglycaemic events ($BG < 2.6$ mmol/L), and hypoglycaemia ($\%BG < 4.0$ mmol/L and $\%BG < 3.0$ mmol/L).
- **PERFORMANCE:** $\%BG$ within the 4.0-8.0 mmol/L band, and hyperglycaemia ($\%BG > 10.0$ mmol/L).

10.3 Results

10.3.1 Comparison of Protocols

Table 10.2 summarises the results for all 4 protocols. Protocol 2 was the most aggressive by design, and thus had higher hypoglycaemia. Protocol 2 has no guarantee of managing hypoglycaemic risk, making it unsuitable for further optimisation despite good performance at higher upper BG limits. In particular, in Figure 10.4, Protocol 2 has a similar distribution slope within the target range to the other protocols, but higher $\%BG$ below 4.0 mmol/L. Protocols 1, 3, and 4 yielded an overall similar performance, which is clearly shown in the overlap of the cumulative distribution curves in Figure 10.4.

Protocol 1, 3, and 4 also performed in similar manner to the NICU-Protocol. This outcome indicates that in most cases, due to large prediction band width in the stochastic model, the NICU-Protocol was putting the 5th percentile of BG outcomes on 4.0 mmol/L to ensure safety, rather than the 50th percentile of BG outcomes on 6.0 mmol/L. This behaviour is seen in the similarities between the percentiles in Protocols 1 and the STAR-NICU protocol in Table 10.2. Thus, the STAR-NICU protocol is implicitly very similar to Protocol 1, even though it was not explicitly designed that way.

The STAR-NICU protocol has 7% greater time in band and fewer episodes of hypoglycaemia (1 of 79 virtual patients vs. 8 of 54 clinical episodes) than the clinical data collected from the clinical use of the STAR-NICU protocol. The STAR-NICU protocol was implemented using the NICU model, previously detailed in Chapter 3, and stochastic models developed from NICU models SI traces. In addition, the stochastic models were based on a much smaller, retrospective, cohort ($N=25$). As such, the performance increase in simulation reflects the change in models more than the protocol itself.

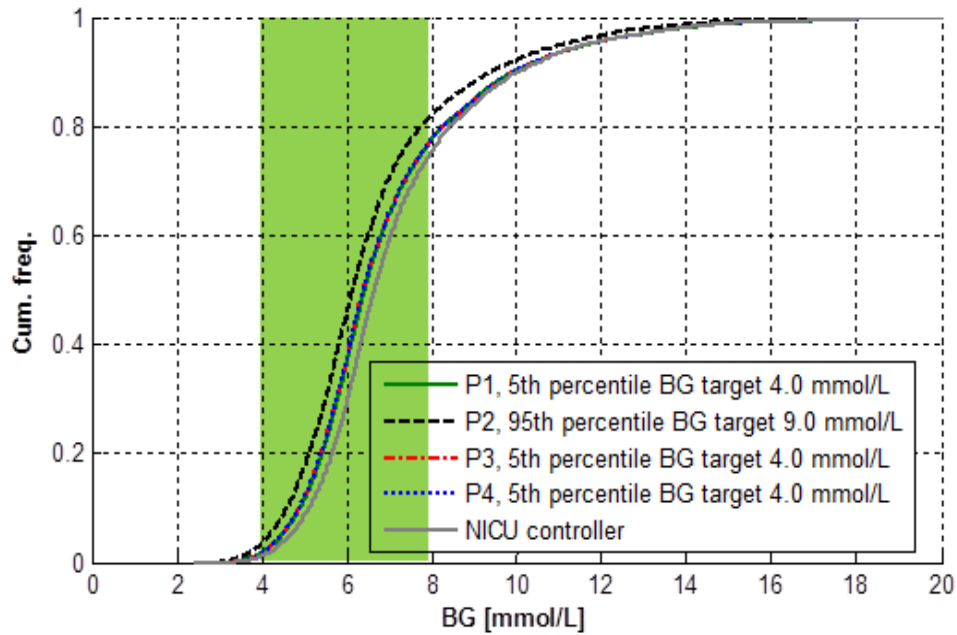


Figure 10.4: Cumulative distributions of BG outcomes for Protocols 1 – 4. Distributions are presented for simulations marked [†] in Table 10.2.

The results for the 5th and 95th percentile targeting approaches of Protocols 1- 4 reveal a typical prediction range of 4.5 – 5.5 mmol/L in size, which is larger than the 4.0 mmol/L width of the target range,. This large width limits the degree to which control to the target range can be achieved. This wide prediction range explains the drop from 14 to 1 hypoglycaemic events in the Protocol 2 simulation results when the limit is raised to 9.0 mmol/L. In this case, for a 95th percentile BG targeted to 9.0 mmol/L, the 5th percentile BG is commonly around 4.0-4.5 mmol/L, giving similar performance to Protocol 1. However, the more aggressive nature of Protocol 2 did not constrain the 5th percentile BG to 4.0 mmol/L or higher, resulting in higher time in band, and higher %BG < 4.0 mmol/L.

Protocols 3-4 showed little gain in preloading insulin to make use of greatest SI certainty, and required more clinical effort in terms of additional insulin interventions between BG measurements. All else equal, reducing workload and expectations also makes a protocol simpler and thus better with greater likelihood of good compliance (Chase et al., 2008b). Thus, Protocol 1 was selected for further optimisation as it balancing desired reductions in hyperglycaemia with reduced hypoglycaemic events and lower clinical effort.

Table 10.2: Virtual trial results for Protocols 1-4

		Performance			Safety			Predictions		
	Target type	Target value	Median BG	% Time in band	%BG>10.0	%BG<4.0	%BG<3.0	# patients with BG<2.6	5th percentile BG predictions	95th percentile BG predictions
NICU protocol clinical data*	50 th 5 th	6.0 4.0	6.6 [5.5 - 8.2]	67.2	11.8	4.6	0.7	8	N/A	N/A
	50 th 5 th	6.0 4.0	6.6 [5.9 – 8.0]	74.9	10.3	1.4	0.1	1	4.5 [4.3 - 4.7]	9.5 [9.1 - 10.1]
Protocol 1		4.0 [†]	6.4 [5.6 - 7.7]	76.1	9.5	2.1	0.2	2	4.3 [4.2 - 4.4]	9.2 [8.9 - 9.8]
		4.2	6.7 [5.8 – 8.0]	73.6	10.7	1.5	0.1	1	4.5 [4.4 - 4.6]	9.6 [9.1 - 10.2]
		4.4	7.0 [6.1 - 8.3]	70.7	11.7	1	0	1	4.7 [4.6 - 4.8]	9.9 [9.5 - 10.7]
		4.8	7.5 [6.6 - 8.8]	62.5	14.8	0.5	0	1	5.1 [5.0 - 5.3]	10.5 [10.0 - 11.2]
		5	7.7 [6.8 - 9.1]	53.4	16.3	0.4	0	0	5.4 [5.2 - 5.6]	10.8 [10.0 - 11.4]
Protocol 2		8	5.6 [4.8 – 7.0]	76.3	8	6.7	0.7	14	3.7 [3.6 - 3.9]	7.9 [7.8 - 8.7]
		9.0 [†]	6.1 [5.3 - 7.3]	78.5	7.9	3.5	0.3	1	4.2 [4.0 - 4.5]	8.9 [8.7 - 9.0]
		10	6.8 [5.9 – 8.0]	73.7	9.4	1.8	0.1	0	4.7 [4.5 – 5.0]	9.8 [9.6 - 9.9]
		11	7.5 [6.5 - 8.6]	62.9	11.5	0.8	0	0	5.3 [5.0 - 5.5]	10.6 [10.3 - 10.8]
		4.0 [†]	6.4 [5.5 - 7.7]	76.4	9.5	2	0.2	1	4.3 [4.2 - 4.4]	9.1 [8.7 - 9.8]
Protocol 3		4.2	6.6 [5.7 - 7.9]	75.2	10.6	1.5	0.1	1	4.4 [4.4 - 4.5]	9.4 [8.9 - 9.8]
		4.4	6.8 [6.0 - 8.1]	72.4	11.8	1.2	0	1	4.7 [4.6 - 4.8]	9.6 [9.2 - 10.1]
		4.8	7.3 [6.4 - 8.6]	65.5	14.2	0.5	0	1	5.0 [5.0 - 5.2]	10.2 [9.7 - 10.9]
		5	7.7 [6.6 - 8.8]	60.7	15.6	0.4	0	0	5.3 [5.2 - 5.5]	10.5 [9.8 - 11.3]
		4.0 [†]	6.4 [5.5 - 7.7]	76.4	9.6	2	0.1	1	4.3 [4.2 - 4.4]	9.1 [8.7 - 9.8]
Protocol 4		4.2	6.6 [5.8 – 8.0]	74	10.6	1.5	0.1	1	4.5[4.4 - 4.6]	9.4 [9.0 – 10.0]
		4.4	6.9 [6.0 - 8.2]	71.3	11.9	1	0	1	4.7 [4.6 - 4.8]	9.7 [9.3 - 10.3]
		4.8	7.4 [6.5 - 8.7]	63	14.9	0.5	0	0	5.1 [5.0 - 5.3]	10.3 [9.8 - 10.9]
		5	7.6 [6.7 – 9.0]	58.5	16.2	0.4	0	1	5.3 [5.2 - 5.5]	10.6 [10.0 - 11.38]

* Clinical implementation of the STAR-NICU protocol used the NICU model and older stochastic models. ** Simulated with the NICING model and new stochastic models. [†] Cumulative distributions are shown for these simulations in Figure 10.4.

10.3.2 Sensitivity Analysis on Protocol 1

Table 10.3 presents a sensitivity analysis on the limit on the increase in insulin and the tolerance on the lower limit. A lower limit of 4.4 mmol/L was chosen such that tolerances of -0.2 and -0.4 mmol/L would allow the insulin rate to be chosen such that the 5th percentile prediction to be within 4.2 – 4.0 mmol/L or 4.0 – 4.4 mmol/L, rather than just greater than a specified lower BG limit.

The limit on the amount by which the insulin rate can increase at any BG level was increased to 0.04, 0.05, and 0.06 U/kg/hr. This change improved time in band, but the incidence of severe hypoglycaemia was increased (results not shown). However, when the amount by which insulin was allowed to increase was varied with BG level this change, in addition to the tolerance on the lower limit, proved effective at increasing the % time in band to around 77%, without increasing the %BG below 4.0 mmol/L. There is some small sensitivity around the BG value that the maximum increase insulin rate is applied. If these BG are lower, time in band is higher, but hypoglycaemia (%BG<3.0 mmol/L) is also higher. A lower tolerance value is theoretically marginally safer, with 1 less incident of severe hypoglycaemia across most of the limit combinations in Table 10.3.

In all simulations the %BG<4.0 mmol/L was lower than a maximum acceptable limit of 5%. In addition, the similar and relatively consistent value of 0.1 % BG<3.0 mmol/L suggests that the 1 extra hypoglycaemic event in the higher tolerance simulations is within simulation noise (Chase et al., 2007, Chase et al., 2010a).

10.3.3 Selected Protocol

The best balance between performance (% time in band) and safety (% incidence of BG<3.0 and number of episodes of hypoglycaemia) was achieved using a tolerance of 0.4 mmol/L, and using BG determined limits on the increase in insulin infusion rate. These limits allow insulin to increase by up to 0.06 U/kg at BG>9 mmol/L, increase by up to 0.03 U/kg/hr between 6.0 and 9.0 mmol/L, and increase by up to 0.01 U/kg/hr below 6 mmol/L. The lowest allowable insulin rate is chosen that results in a 5th percentile BG prediction within the tolerance range of 4.0 – 4.4 mmol/L. If the 5th percentile BG is higher than this tolerance range with the maximal allowable insulin rate, then this maximum insulin rate is given. This optimised protocol will be denoted as the STAR-GRYPHON (STAR glycaemic regulation system to prevent hyper- and hypo- glycaemia in neonates) protocol.

Table 10.3: Virtual trial results for the optimisation of Protocol 1.

	Tolerance	BG dependent rate increase	Median BG	% Time in band	%BG>10	%BG<4.0	%BG<3.0	# patients with BG<2.6
Targeting type: 5th on 4.4 mmol/L	-0.2	5.0, 8.0	6.6 [5.8 - 7.8]	75.6	9.3	1.5	0.1	1
		6.0, 9.0	6.4 [5.6 - 7.7]	74.4	9.5	1.4	0.1	1
		6.5, 9.0	6.7 [5.8 - 7.9]	73.8	9.8	1.4	0.1	0
		6.0, 10.0	6.5 [5.6 - 7.7]	73.8	9.8	1.3	0.1	0
		7.0, 10.0	6.7 [5.9 - 8.0]	73.4	9.9	1.3	0.0	0
		7.0, 11.0	6.5 [5.7 - 7.8]	73.3	10.2	1.2	0.0	0
	-0.4	5.0, 8.0	6.7 [5.9 - 8.0]	77.6	8.5	2.3	0.2	3
		6.0, 9.0	6.5 [5.7 - 7.8]	76.4	9.0	1.8	0.1	2
		6.5, 9.0	6.8 [5.9 - 8.0]	75.6	9.2	1.8	0.1	1
		6.0, 10.0	6.6 [5.7 - 7.9]	75.5	9.3	1.7	0.1	1
		7.0, 10.0	6.8 [5.9 - 8.0]	75.1	9.6	1.6	0.0	1
		7.0, 11.0	6.6 [5.7 - 7.9]	74.7	9.9	1.6	0.0	1

10.4 Discussion

Virtual trials have been used to optimise a STAR protocol, leading to the STAR-GRYPHON protocol for use in the NICU. Virtual trials using virtual patients derived from clinical data enable different protocols to be tested quickly and safely, without the undue cost and risk of extended clinical trials. This new protocol uses stochastic targeting to place the 5th percentile of predicted BG outcomes within tolerance of the lower BG limit of 4.4 mmol/L to maximize overlap between the range of possible BG outcomes and a clinically specified target band (4.0-8.0 mmol/L). Further, the maximum increase in insulin is limited based on BG level, so that as the risk of hypoglycaemia decreases, more aggressive insulin treatments can be utilised. This approach allows direct management, of both hyper- and hypo- glycaemic risk.

Performance is better than the NICU-Protocol in simulation by 1.7%, which is lower than might be expected. However, due to the large band width resulting from the stochastic model, the NICU-Protocol puts the 5th percentile of BG outcomes on 4.0 mmol/L to ensure safety, rather than the 50th percentile of BG outcomes on 6.0 mmol/L. As a result the protocols are

similar in effect, with the STAR-GRYPHON protocol's advantage being in its relative simplicity and further optimisation for this cohort.

In virtual trials, the STAR-NICU protocol has higher time in band and fewer episodes of hypoglycaemia than the clinical data collected from the clinical use of the STAR-NICU protocol. This performance increase reflects the change in model, as the previous STAR-NICU protocol was implemented using the NICU model of Chapter 3, and different stochastic models based on the NICU model and less data. This relative increase in both performance and safety indicates that the NICING model is more effective at capturing relevant insulin-glucose physiology for the purposes of glycaemic control, thus narrowing stochastic forecasting bounds. It may also reflect the larger clinical population that the new stochastic models are based on.

The %BG below 4.0 mmol/L was much lower than the theoretical maximum of 5%. This outcome reflects the conservative nature of population based models for estimating patient-specific changes in SI. However, this conservativeness increases safety from hypoglycaemia, as many patients do not drop below 4.0 mmol/L.

The effect of patient variability can be represented by the width of the 5th - 95th percentile range of likely BG outcomes, which is given for various protocols in Table 10.2. This range is usually 4.5-5.5 mmol/L in width, which is larger than the width of the target range. The ability to control tightly to this target range is therefore partially compromised. The width of this 5th-95th percentile BG outcome range is larger at higher blood glucose levels, where wide prediction bands in S_I are magnified by high BG and large recommended insulin dosage (see Equation 9.9).

Limits on the maximum change in insulin rate enable more aggressive treatments to be given at higher BG. However, they also provide an improved degree of safety at lower BG, as allowable insulin rates are more restricted. This restriction helps moderate risk of hypoglycaemia due to sudden rises in SI when at low BG, as well as from over response to any significant measurement error which can occur.

The slightly higher incidence of hypoglycaemia seen in Table 10.3 was due to sensitivity to a sudden large increase in virtual patients SI while the patient was at a BG of ~8.0 mmol/L or higher. In most cases these large SI spikes occur around large changes in the parenteral nutrition rate in the original clinical data. These spikes are likely unphysiological model

artefacts, and reflect error around the onset of these nutrition rate changes, delays due to dead space in the delivery tubing, or inability of the model to accurately capture rapid changes in BG due to these nutrition changes.

Clinical limitations on performance are present in the form of restrictions on the number of BG measurements that can be taken in a set time period. Neonates have extremely limited blood volume, so the lower limit on measurement interval is 2 hours, with clinical preference being 3-4 hours and a preferred maximum of around 8 measurements per day. Hence, the control protocols are optimised for 4 hourly intervals, although 3 hourly intervals give better performance.

10.5 Summary

Virtual patients were created using clinical data, and were used in virtual trials to test and optimise a protocol for dosing insulin in very/extremely premature infants. The STAR-GRYPHON protocol was developed, which uses stochastic targeting of predicted SI outcomes to generate a range of BG predictions for a given insulin dose. The protocol selects and insulin infusion rate such that the maximum possible likelihood of BG outcomes less than 4.0 mmol/L is 5%. Limits on the amount that the insulin infusion rate can increase between treatments are applied according to current BG, allowing more aggressive treatments when the current BG is above 9.0 mmol/L.

Chapter 11: Clinical results

In Chapter 10 virtual trials were used to develop and optimise a protocol in silico. The final optimised protocol is known as STAR-GRYPHON. Chapter 11 presents clinical results from the use of the STAR-GRYPHON protocol in Christchurch Women's Hospital NICU.

11.1 Introduction

In this Chapter clinical results are presented from patients treated using the new NICING model summarised in Chapter 9 and the STAR-GRYPHON protocol developed in Chapter 10 are presented. These results are compared to clinical results from previous work by (Le Compte et al., 2009). This previous work included a protocol, denoted here as the STAR-NICU protocol that uses the NICU model previously described in Chapter 3.

11.2 Methods

11.2.1 Model Dynamics and Stochastic Forecasting

The glucose-insulin metabolic models utilised by the STAR-GRYPHON protocol and the older STAR-NICU protocol were the NICING and NICU models, respectively. These are summarised in Chapters 9 and 3. In the case of the NICING model, the premature infant specific parameters for the gut model were not yet developed, so the adult ICU values of $d_1 = 0.035 \text{ min}^{-1}$ and $d_2 = 0.007 \text{ min}^{-1}$ (corresponding to half lives of 20min and 100min respectively) were used with the 2 compartment gut model.

Likely future outcomes in SI were forecast in both protocols using the stochastic modelling method outlined in Chapter 3. Stochastic models were created based on clinical data from the retrospective cohort in the case of the STAR-NICU protocol, and the retrospective and STAR-NICU clinical cohorts in the case of the STAR-GRYPHON protocol.

11.2.2 Protocol Definition

STAR-NICU Protocol: Target to Value

The 50th percentile of likely SI outcomes is used to determine the 50th percentile of BG outcomes for defined insulin and nutrition inputs. An insulin rate is chosen, such that the 50th percentile of BG outcomes is placed on a median target of 6.0 mmol/L and the 5th percentile of likely BG outcomes is greater than 4.0 mmol/L (Le Compte et al., 2009) (see Figure 11.1a). If the second condition is not met, insulin is reduced until it is, and the 50th percentile is allowed to be higher than the median target of 6.0 mmol/L. As added protection against noise or mis-measurement, the rate of change of insulin is limited to a maximum increase of 0.03 U/kg/hr per treatment, but can be dropped to zero if required. The overall approach was designed to balance performance and hypoglycaemic risk relatively equally. As a target to value protocol it is similar to many clinical protocols, but with the added model-based assessment and management of risk.

STAR-GRYPHON Protocol: Target to Range

To determine baseline SI the infant is started on 0.05 U/kg/hr insulin infusion for 2 hours. From there, the protocol used clinically was the optimised STAR-GRYPHON protocol developed in Chapter 10. To summarise briefly, an allowable insulin rate is chosen such that the 5th percentile of BG outcomes is placed at the maximum possible value between 4.0- 4.4 mmol/L, as shown in Figure 11.1. Adaptive limits on the amount by which the insulin rate is allowed to increase are applied. If current BG is greater than 9.0 mmol/L, insulin infusion can increase by up to 0.06 U/kg/hr. If BG is between 6.0-9.0 mmol/L it can increase by up to 0.03 U/kg/hr, and if BG<6 mmol/L it can only increase by up to 0.02 U/kg/hr. Insulin infusion rates are allowed to decrease or be turned off at any time.

11.2.3 Clinical Cohorts

Data from hyperglycaemic (2 consecutive BG \geq 10.0 mmol/L within 4-6 hours) premature neonates (GA<30 weeks, BW<1.5 kg) treated with insulin in Christchurch Women's Hospital NICU was obtained for three cohorts: 1) Retrospective bedside titration (pre August 2008); 2) the first STAR-NICU protocol (August 2008-December 2012); and 3) the optimised STAR-GRYPHON protocol (January 2013-June 2014). Pilot trials under the STAR-NICU protocol were approved by the Upper South A Regional Ethics Committee, New Zealand

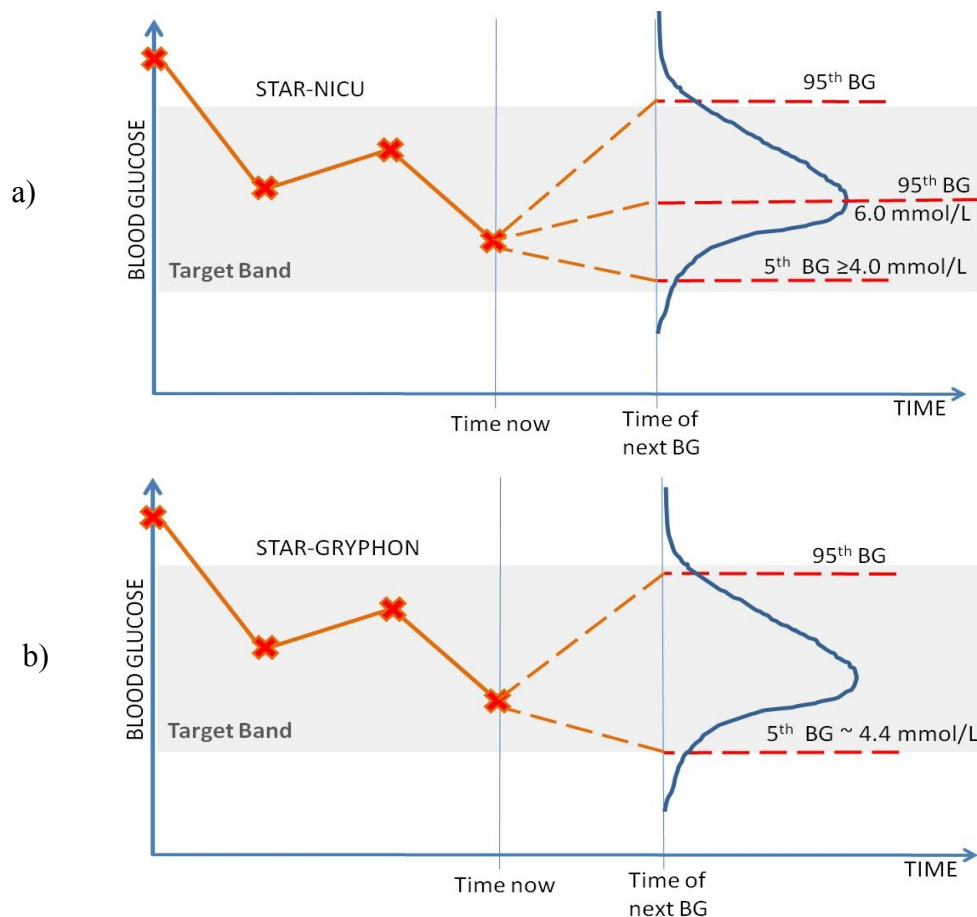


Figure 11.1: Dosing protocols use the forecast ranges of likely BG outcomes to select an appropriate insulin dose. The STAR-NICU protocol (a) targeted the outcome median to 6.0 mmol/L and checked that 95% of likely BG outcomes were greater than 4.0 mmol/L, whereas the STAR-GRYPHON protocol (b) dosed insulin such that 95% of likely outcomes were greater than 4.4 mmol/L.

(ClinicalTrials.gov registration NCT01419873). From August 2010 onwards, STAR was implemented clinically as a standard of care. Glycaemic data was de-identified and stored automatically by the computer system for data audits on performance and safety, and to increase the population data set that the statistical models draw on.

From January 2013 to June 2014, 13 premature infants totalling 16 episodes of insulin therapy received insulin under STAR-GRYPHON. The STAR-NICU cohort comprised of data from 40 premature infants totalling 61 episodes, who received insulin therapy under the STAR-NICU protocol, as either a pilot trial or as a standard of care, from August 2008 to December 2012. The first 22 patients in this cohort have been previously reported (Le Compte et al., 2012). Retrospective data was obtained for 22 patients, comprising 25 glycaemic episodes, who received insulin under a bedside titration protocol prior to STAR implementation in 2008. Cohort characteristics are included in Table 11.1 in the results.

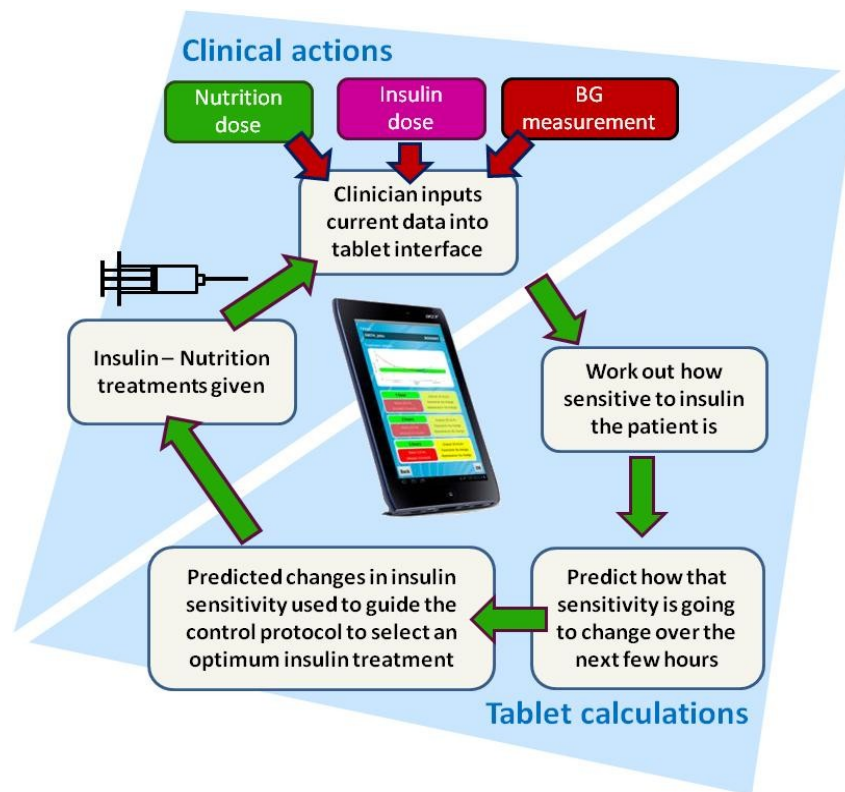


Figure 11.2: Clinical implementation of STAR glycaemic control. Clinical glycaemic control actions are used by the tablet to calculate and predict future changes in a patient’s insulin sensitivity. Insulin doses are then chosen based on likely future outcomes in blood glucose.

A patient episode was defined as 12 hours or more of insulin therapy with at least 4 BG measures. Episodes are separated by at least 12 hours reflecting separate episodes of treatment and a full restart of the glycaemic control protocol. STAR was initially implemented using a PC computer based application, and more recently through the use of bedside tablet computers with a touch-screen interface.

11.2.4 Clinical Implementation

The clinical implementation cycle for the STAR protocol is shown in Figure 11.2. STAR-GRYPHON was implemented as a tablet computer (Samsung Galay Tab 2, 10.1 inch) application with a specifically designed graphical user interface, part of which is shown in Figure 11.3. The user interface, calculations, and data storage were all coded in Java and implemented using the android operating system (Android version 4.0.4). Solver and forward simulation Java code was structured in a similar manner to the Matlab[®] code used in virtual trials. A 4th order Runge-Kutta ODE solver was written in java to mimic the action of ode45 solver in Matlab[®], which it did to 4 or 5 decimal places. Matlab simulations calling the Java

code for the protocol and model solvers were carried out to ensure consistency between simulation and real-world implementation.

The interface (see Figure 11.3) was set up to ensure clarity of current clinical inputs and confirmation of treatments given to minimise clinical error effects on insulin dose calculation. Insulin and nutrition records are reviewed and/or updated prior to the insulin dose calculation. This data and the current BG measure are used to calculate the current value of SI and recommend an insulin dosage based on SI forecasts using this value, as illustrated in Figure 11.1. Having recommended an insulin dosage, at the end of the measurement interval the next BG measurement is entered into the tablet, alongside any changes in nutrition or insulin, and another treatment is calculated.

A neonate is taken off STAR when BG is in the target range (4-8 mmol/L), no insulin has been given for 6 hours, and/or the infant is determined to be glycaemically stable. An infant may be restarted as a new episode if hyperglycaemia recurs.

Blood samples are taken from a central umbilical line, if available, or from heel pricks every 2-4 hours. BG is determined using a Bayer 850 blood gas analyser (Bayer AG, Leverkusen, Germany). Insulin is always delivered as an infusion through the same line as the main dextrose fluids. Insulin concentration is calculated based on infant weight, such that $0.05 \text{ U/kg/hr} = 0.20 \text{ mL/hr}$. Infusions are given in steps of 0.01 U/kg/hr , delivered by the pump in to an resolution of 0.01 mL/hr . Delivery tubing is pre-soaked in the insulin solution for 15 minutes to reduce the effect of insulin absorption by the tubing on glycaemic control.

Nutrition rates are clinically determined, and are not normally adjusted for glycaemic control. All recent (last 3-4 hours) and planned future nutrition sources and rate changes are accounted for when determining insulin dosage. Typically, neonates receive the bulk of their nutrition via TPN with 10% dextrose, with a target total glucose delivery of 6-8mg/kg/min. Medications, such as dobutamine and morphine, may be infused with 5% dextrose or saline solutions, which are also accounted for in the model calculation. Enteral boluses of expressed breast milk (EBM) are used as soon as they are available, starting at 0.5mL every four hours, with increasing volume and frequency as tolerated. Milk formula may also be given enterally, and human milk fortifiers were sometimes added to EBM to increase caloric value. All this information was available to the model at the time of calculation.

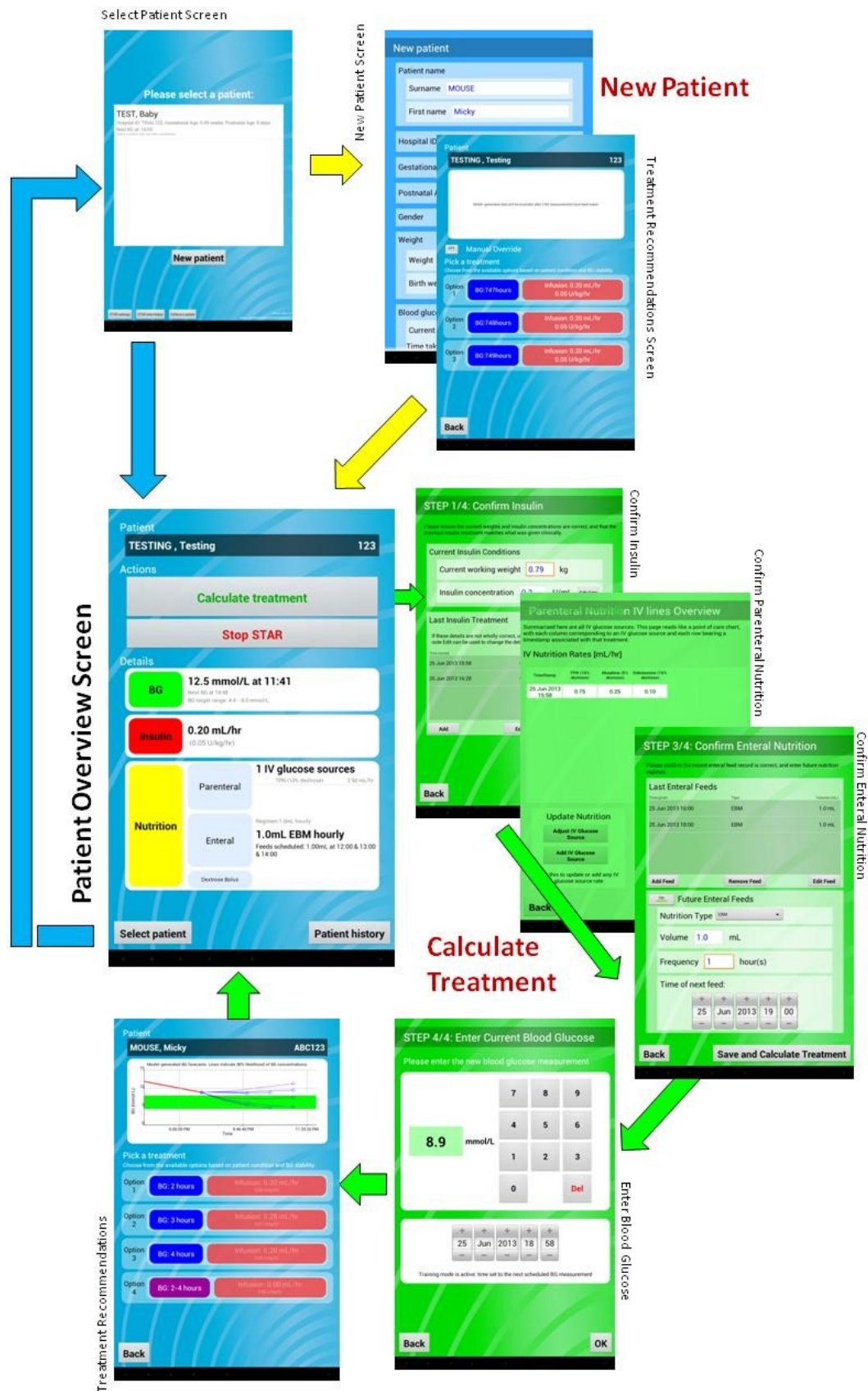


Figure 11.3: STAR-GRYPHON graphical user interface for treatment calculation.

11.2.5 Analysis

Model BG data is re-sampled hourly because where the measurement interval is different, re-sampled statistics allow a fairer comparison between groups, as well as an indication of glycaemic behaviour between measurements. Re-sampled statistics use the model generated BG profile to estimate BG behaviour between measurements.

Safety is evaluated by the number of patients with one or more hypoglycaemic events ($BG < 2.6$ mmol/L) and the %BG below the lower target band value of 4.0 mmol/L. Performance is determined by the percentage time in a 4.0-8.0 mmol/L target band, and minimisation of hyperglycaemia by the %BG > 10.0 mmol/L.

Statistical comparisons of median values are made using the Mann-Whitney U test, and the Kolmogorov-Smirnov test to compare variability (Matlab[®], ranksum, kstest2). The Fishers exact test was used to calculate the statistical significance of the number of patients with at least 1 hypoglycaemic episode. Statistical power, based on sample size for $p=0.05$, is calculated using the method presented in Whitley and Ball (2002). Statistical power was considered for 3 metrics: 1) per-patient percentage time in targeted range; 2) per-patient median BG, and 3) number of patients with hypoglycaemia. Consideration was taken of unequal cohort sized in comparison.

11.3 Results

11.3.1 Clinical Results

Performance statistics for each cohort are given in Table 11.1. The STAR-GRYPHON protocol implementation saw re-sampled BG in the target band rise to 76.6% (72% of all BG measures), with no incidence of hypoglycaemia ($BG < 2.6$). This outcome reflects an almost 10% absolute increase in BG in the target band in comparison to STAR-NICU, and a 26% absolute increase with respect to retrospective data. Statistical power for percentage time in band statistics was >95% and 88% respectively for STAR-GRYPHON and STAR-NICU cohort comparisons to retrospective data.

Median BG was lower for STAR-NICU and STAR-GRYPHON compared to retrospective data (6.6 & 6.9 mmol/L vs. 7.8 mmol/L, $p=0.01$ (power 74%) and $p=0.09$ (power 30%), respectively). Cumulative distributions of BG in Figure 11.4 are significantly different ($p < 0.005$ for all comparisons), and are steeper for STAR-NICU and STAR-GRYPHON in

comparison to retrospective data. While STAR-GRYPHON had higher median BG than STAR-NICU, it had the steepest gradient, indicating the tightest range of clinical glycaemic outcomes. Notably, STAR-GRYPHON had much less BG<4.0 mmol/L (p=0.007 in comparison to retrospective data, and p<0.001 in comparison to STAR-NICU), with no incidence of hypoglycaemia. That there were no patients who experienced hypoglycaemia on the STAR-GRYPHON protocol was not statistically significant in comparison to STAR-NICU (p=0.14) given the smaller patient numbers. For 80% statistical power and p=0.05 a total of at least 26 STAR-GRYPHON patients would be required.

Table 11.1: Clinical results and glycaemic statistics by cohort. Data are given as Median [Inter-quartile range].

	Retro- spective (R)	STAR- NICU (SN)	STAR- GRYPHON (SG)	P value		
				SN vs. R	SG vs. R	SG vs. SN
n Patients (n male)	22 (7)	40 (22)	13 (8)			
n Episodes	25	54	16			
Total Hours	3098	5014	987			
Number of BG measurements	943	1632	270			
Gestational Age (GA) [weeks]	27 [25-27]	26 [25-29]	26 [26-27]	0.82	0.91	0.84
Weight [g]	845 [800-900]	785 [635-950]	908 [820-1030]	0.66	0.24	0.25
Post Natal Age (PNA) [days]	3 [1-7]	3 [1-12]	3 [2-4]	0.42	0.94	0.45
Measurement interval	3.2 [2.6 - 3.9]	3.3 [2.7 - 3.8]	4.0 [3.8-4.1]	0.98	0.01	<0.001
Insulin rate [IQR] (U/kg/hr)	0.03 [0.02-0.06]	0.04 [0.02-0.08]	0.05 [0.04 - 0.07]	0.06	0.002	0.02
Total glucose delivery rate [IQR] (mg/kg/min)	8.1 [5.7 - 9.7]	7.3 [5.1- 8.5]	7.7 [7.1 - 8.6]	0.50	0.81	0.84
<i>Hourly re-sampled statistics</i>						
Median BG	7.8 [6.6- 9.1]	6.6 [5.5 - 8.2]	6.9 [6.1-7.9]	<0.001	<0.001	<0.001
% BG within 4.0 - 8.0 mmol/L	50.3	67.2	76.6	<0.001	<0.001	0.42
% BG>10 mmol/L	17.7	11.8	5.6	0.01	0.005	0.42
% BG<4.0 mmol/L	3.0	4.6	0.4	0.21	0.007	<0.001
% BG<3.0 mmol/L	0.3	0.7	0	0.28	0.09	0.02
Num episodes with BG <2.6 (%)	1 (4)	8 (13)	0 (0)	0.20	0.61	0.14

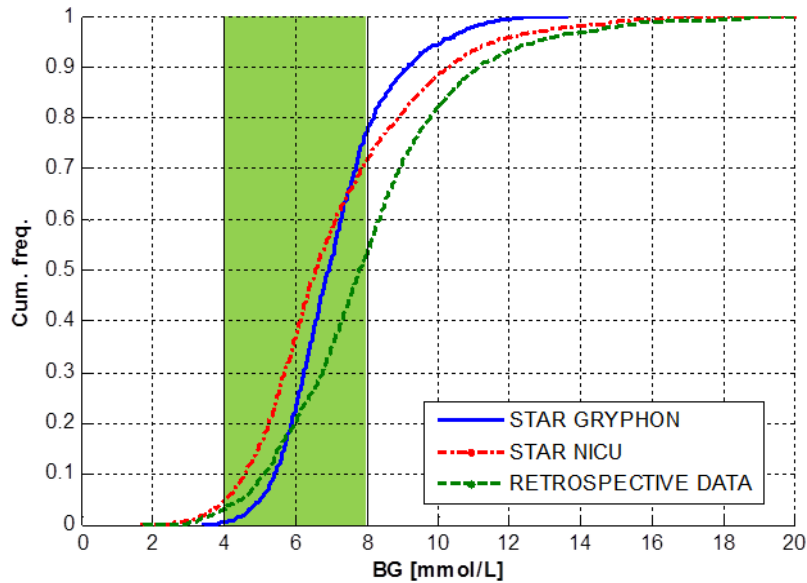


Figure 11.4: Blood glucose (BG) distributions for each patient cohort. The vertical axes denotes the proportion of BG measurements \leq the given horizontal axis threshold, and the clinically targeted range is shaded.

11.3.2 Comparison to Simulation

The STAR-GRYPHON shows similar performance in clinical results to those predicted in simulation, with 76.6 vs. 76.4% time in band, respectively. However, the incidence of %BG < 4.0 mmol/L is significantly lower than predicted in simulation ($p=0.03$). The incidence of hypoglycaemia (number of patients with at least 1 BG < 2.6 mmol/L) and hyperglycaemia (%BG > 10.0 mmol/L) is also lower than predicted in simulation, although not significantly so ($p \geq 0.60$). The cohort characteristics, such as weight, GA, and PNA are also similar ($p \geq 0.16$), as well as clinically delivered insulin and glucose. The median BG is higher in the clinical results than in simulation. The median measurement interval is the same as simulated.

11.3.3 Per-patient Results

Individual patient results for STAR-GRYPHON are given in Table 11.3. Performance varies between patients, with a general trend of increased performance with GA, but not necessarily BW. Patients 1 and 10 are very resistant to insulin to start with. The results for Patient 1, shown in Figure 11.5a, show that BG is greater than 7.5 mmol/L for the first 55-60 hours of glycaemic control, despite relatively high insulin doses. Insulin dose is still lower than the clinically imposed maximum dose of 0.3 U/kg/hr, as the BG prediction bands for this patient are wide, with the actual BG closer to the cohort-based 95th percentile than the median. Hence, slightly higher BG results from wide stochastic model bounds at high insulin doses, and provides additional safety from hypoglycaemia.

Table 11.2: Virtual trial vs. Clinical results for the STAR-GRYPHON protocol. Data are given as Median [Inter-quartile range].

	Virtual Trial results	Clinical results	P value
n Patients (n male)	62 (29)	13 (8)	
n Episodes	79	16	
Total Hours	7962	987	
Number of BG measurements	2109	270	
Gestational Age (GA) [weeks]	26 [25-28]	26 [26-27]	0.99
Weight [g]	845 [635 – 940]	908 [820-1030]	0.16
Post Natal Age (PNA) [days]	3 [1-8]	3 [2-4]	0.57
Measurement interval	4.0	4.0 [3.8-4.1]	-
Insulin rate [IQR] (U/kg/hr)	0.05 [0.03 - 0.08]	0.05 [0.04 - 0.07]	0.80
Total glucose delivery rate [IQR] (mg/kg/min)	7.5 [5.4 - 9.2]	7.7 [7.1 - 8.6]	0.70
<i>Hourly re-sampled statistics</i>			
Median BG	6.5 [5.6 - 7.7]	6.9 [6.1-7.9]	<0.001
% BG within 4.0 - 8.0 mmol/L	76.4	76.6	0.44
% BG>10 mmol/L	9.0	5.6	0.60
% BG<4.0 mmol/L	1.8	0.4	0.03
% BG<3.0 mmol/L	0.1	0.0	0.25
Num episodes with BG <2.6 (%)	2	0 (0)	0.69

A more typical patient is shown in Figure 11.5b. In this case BG is lowered quickly and remains in band for a high proportion of the remaining time. Insulin is continually modulated, even when BG is within band, in response to changing SI. Insulin is eventually turned off as SI rises.

Compliance with the STAR-GRYPHON protocol was high. Over 16 patient episodes (totalling 272 treatments) there were 10 treatment instances of changes to the STAR-GRYPHON protocol recommendations (3.6% of all treatments). Once (10%) insulin was given when it was recommended that it be turned off, and in 3 (30%) instances insulin was changed independently. In 6 instances (60%), insulin was overridden and turned off. In this last case, insulin was turned off in 4 instances (40%) at the end of a patient episode instead of maintaining the minimum allowable insulin infusion. This occurred twice consecutively (20%) within the same patient at a BG of 8.3 and 8.9 mmol/L, respectively, without further explanation. Compliance data was not available for the STAR-NICU protocol.

Table 11.3: Individual results and protocol compliance across the STAR-GRYPHON cohort.

Patient	Total hours (# BG measures)	Initial BG [mmol/L]	% BG>10 [mmol/L]	% 4.0<=BG<=8.0 [mmol/L]	# BG<4 [mmol/L]	# BG<3.5 [mmol/L]	# Hours till<8.0 [mmol/L]	% 4.0<=BG<=8.0 [mmol/L] after first BG<8.0 mmol/L	# non-compliance
1	96 (28)	15	9.8	56.5	0	0	10.3	64.2	2
2	91 (22)	11.1	4.4	87.9	0	0	2.9	90.9	0
3	25 (9)	10.7	3.8	69.2	1	0	4.2	85.7	0
4	18 (6)	12.6	10.5	63.2	0	0	4.6	85.7	2
5	88 (24)	8.0	0.0	90.4	0	0	N/A	91.7	0
6 Ep. 1	67 (17)	11.4	6.3	75.0	0	0	0.6	76.2	0
6 Ep. 2	24 (7)	11.3	20.0	60.0	0	0	0.9	62.5	1
7 Ep. 1	29 (9)	12.0	10.3	58.6	0	0	6.3	77.3	2
7 Ep. 2	58 (15)	11.6	1.7	79.7	1	0	8.3	94.0	1
8	243 (66)	10.5	3.3	85.7	0	0	0.8	86.0	0
9	63 (17)	10.2	3.1	78.1	0	0	2.7	82.0	0
10	111(28)	11.6	9.9	58.6	0	0	19.5	71.4	1
11	71 (17)	12.6	7.0	80.3	0	0	4.9	86.4	0
12	50 (13)	10.9	3.1	88.0	0	0	6.8	91.7	0
13 Ep. 1	19 (6)	11.7	18.2	54.5	1	0	3.2	85.7	0
13 Ep. 2	20 (7)	11.8	5.0	90.0	0	0	0.2	94.7	1

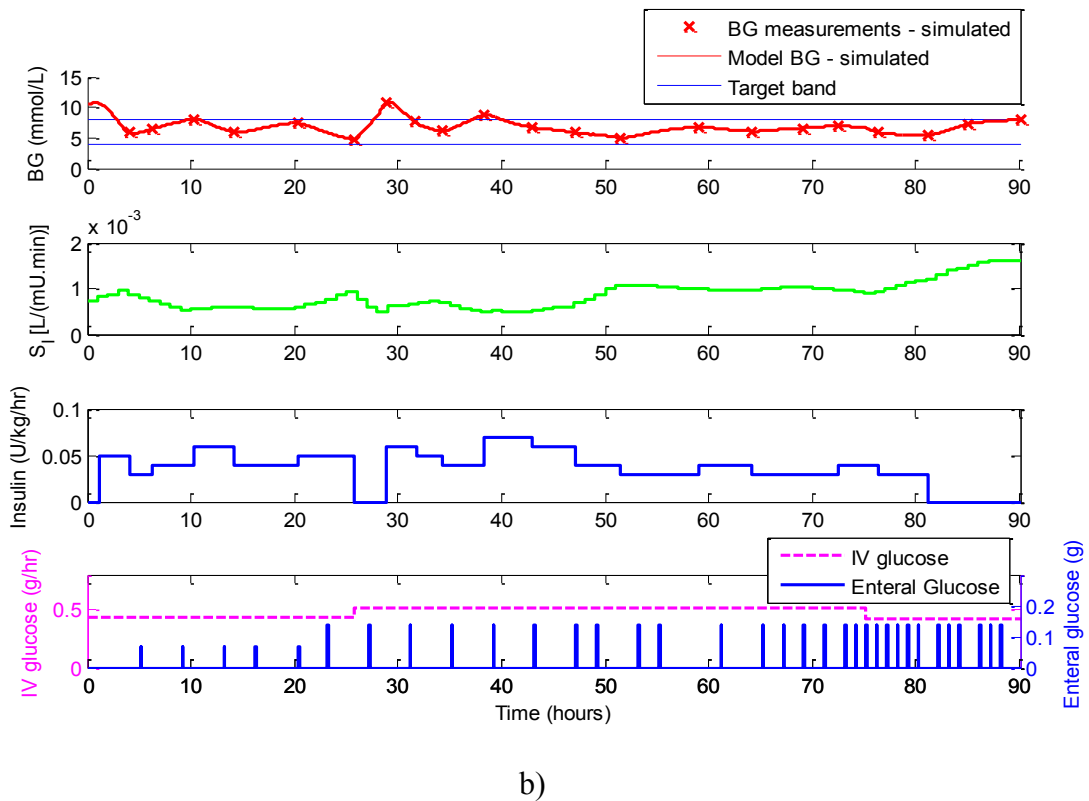
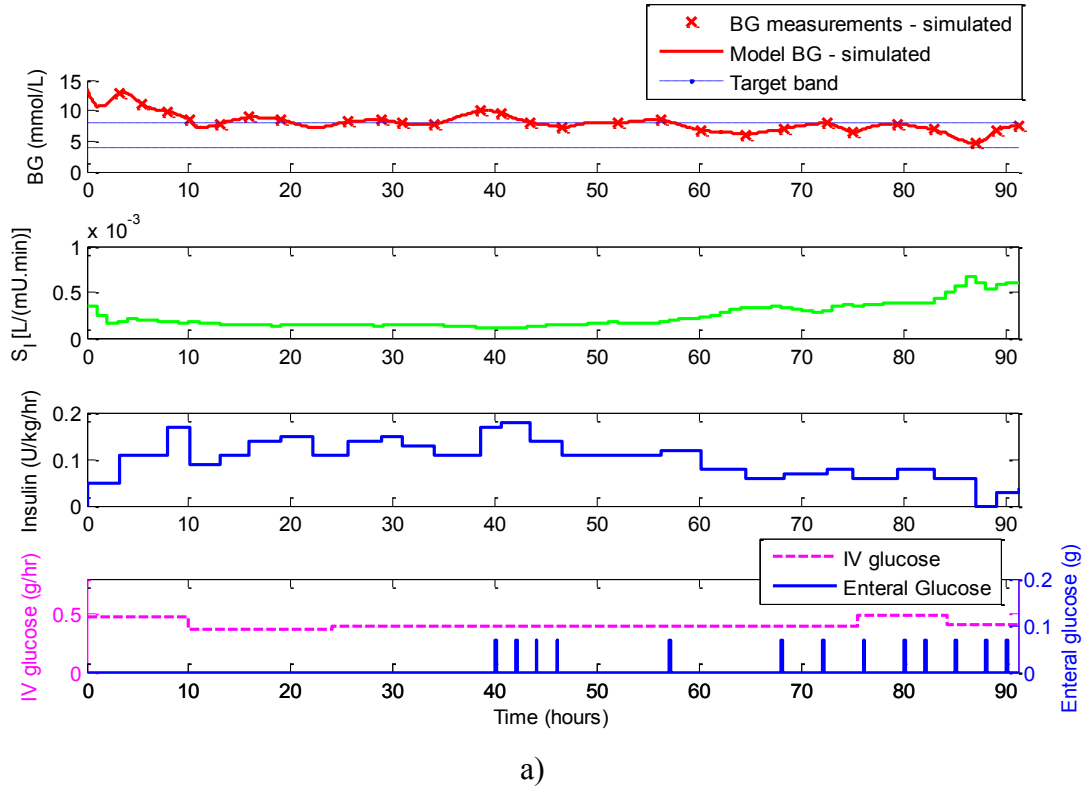


Figure 11.5: STAR-GRYPHON clinical results for a) Patient 1, and b) Patient 2. Plots show blood glucose (BG), model-based insulin sensitivity (SI), and clinical insulin and nutrition treatments.

11.4 Discussion

11.4.1 Protocol Performance

The STAR-NICU and STAR-GRYPHON protocols increased the percentage of BG measurements within the target band of 4.0 – 8.0 mmol/L in comparison to retrospective data. In the case of the STAR-NICU protocol this result was predominantly due to the reduction of hyperglycaemia and lowering of the median patient BG, and results across an increased cohort of patients reflect those of early published pilot trials (Le Compte et al., 2012). STAR-GRYPHON improves on the performance of STAR-NICU by further reducing hyperglycaemia, and significantly reducing the incidence of BG < 4.0 mmol/L ($p < 0.001$). In addition, clinical workload, as reflected by the BG measurement interval, was reduced. Under the STAR-GRYPHON protocols there were no (0%) patients with at least one incidence of hypoglycaemia, but this was not statistically significant due to lower patient numbers. Figure 11.4 shows a significantly steeper cumulative distribution of BG ($p < 0.005$), indicating much tighter glycaemic control with greater safety from hypoglycaemia than either retrospective clinical data or STAR-NICU could achieve.

A key difference in protocol between STAR-GRYPHON and STAR-NICU is a shift from targeting a median target BG to instead optimise the overlap of likely outcomes with a target band. In addition, adapted limits on the allowable increase in insulin infusion rate are applied, with greater restrictions in this increase at lower BG. In practice, high variability means that both protocols are very similar, except in their treatment of BG near the lower end of the normal range, where results show STAR-GRYPHON to be safer and more effective. This result was previously seen in simulation, and is discussed in Chapter 10. The greater difference in the percentage time in band in clinical results compared to the simulation results of Chapter 10 (9 vs. ~2%) reflects the contribution of the new NICING model to the tightening of control.

The average delivery rate of insulin is higher under STAR-GRYPHON, while the median glucose rate was highest in the retrospective data. The glucose delivery rate was also most varied in the retrospective data, suggesting that to some extent nutrition was being modulated to control BG levels at that time. Glucose delivery rates are determined clinically, and were lower for patients under STAR-NICU and STAR-GRYPHON, but not statistically different. However, the lower median BG cannot solely be attributed to lower glucose delivery rate, as average insulin delivery rates are higher. In addition, Chapter 10 shows that for the same

glucose delivery rates given clinically in the retrospective cohort, STAR-GRYPHON was expected to lower the median BG and increase the time in band, thus tightening control. Higher insulin rates, improved control, and lack hypoglycaemic incidences reflect STAR-GRYPHON's ability to modulate insulin according to glycaemic response and overall patient condition.

11.4.2 Comparison to Simulation

The clinical performance of STAR is similar to what was predicted in simulation, with a percentage time in band of 76.6 in comparison to 76.4 ($p=0.44$). However, the $\%BG<4.0$ is much lower ($p=0.03$). The lower incidence of hyperglycaemia suggests overall higher whole body SI than predicted by the stochastic models. In addition, lower hypoglycaemia indicates additional safety, and suggests that the stochastic models over predict both extremes in the clinical patients treated with STAR-GRYPHON to date.

The median BG is lower in simulation than in the clinical results (6.5 vs. 6.9 mmol/L, $p<0.001$). This is due in part to the clinical tendency near the end of a glycaemic episode to override and turn insulin off, rather than maintain a minimum insulin rate, as notes in Section 11.3.3. It could also reflect the conservative nature of stochastic forecasting in this cohort.

11.4.3 Comparison to Literature Results

As discussed in Chapter 2, the benefits of insulin therapy to promote increased glucose tolerance and weight gain have been well reported (Binder et al., 1989, Collins et al., 1991, Kanarek et al., 1991, Meetze et al., 1998, Ostertag et al., 1986, Pollak et al., 1978, Thabet et al., 2003, Vaucher et al., 1982), but few statistics are reported with respect to the quality of glycaemic control. Rates of hypoglycaemia, when discussed, are often reported as the percentage of all BG measures taken, and are typically less than 1% $BG<2.2$ or 2.6 mmol/L. Further, target ranges and definitions of hyper- and hypo- glycaemia vary across centres (Alsweiler et al., 2007) and between studies. Thus, very little literature is available with which these STAR glycaemic control results can be compared. Such difficulties are further compounded by differences in practice and definition. However, limited comparisons can be made to the studies previously outlined in Chapter 2, Section 2.3.

The HINT trial randomised control trial saw a doubling of the number of incidences of hypoglycaemia (58% of infants with one or more $BG<2.6$ mmol/L in the TGC group compared with 27% in the control group) is reported, despite the fact that around a third of

the control group ended up being treated with insulin as well (Alsweiler et al., 2012). Beardsall and colleagues (Beardsall et al., 2008) report a high rate of hypoglycaemia (defined as BG<2.6 mmol/L for more than 60 min when measured by continuous glucose monitor (CGM)), with 29% of infants receiving early insulin therapy experiencing one or more hypoglycaemic events compared with 17% in the control group. In a study comparing insulin therapy between ELBW and LBW infants, Ng and colleagues (Ng et al., 2005) report that 15% of ELBW and 11% of LBW infants had BG<2.6 mmol/L during their study. In comparison, in the STAR-NICU cohort 8/62 (13% of all episodes) had hypoglycaemic events, and STAR-GRYPHON to date has shown no (0 %) hypoglycaemic events, and no BG<3.5 mmol/L.

The median BG from neonatal STAR was 6.6 [5.5 - 8.2] mmol/L for STAR-NICU and 6.9 [6.1 - 7.9] mmol/L for STAR-GRYPHON, which is higher than the 6.2±1.4 mmol/L seen in the early insulin group of the NIRTURE trial and higher than the HINT TGC group median of mean daily glucose measure 5.8 [4.8-6.7] mmol/L. It should be noted that the glycaemic control statistics are only reported for study days 2-7 in the NIRTURE trial, ignoring earlier, potentially higher and more unstable BG values that can occur early in the stay. Comparison to Ng is made difficult as only the median [range] BG during insulin therapy is reported (12 [6-24] mmol/L in the ELBW cohort, and 11 [7-19] mmol/L in the LBW cohort), which are much higher than those of STAR.

11.4.4 Strengths and Limitations

Compliance with STAR-GRYPHON was high (96.4%), and 4 of 10 of non-compliance events (40%) were due to insulin being turned off when an infant was judged glycaemically stable near the end of an episode, instead of maintaining a minimum insulin rate. Another source of non-compliance was insulin rates being dropped between BG measures, in most cases because of a drop in nutrition rate. While, ideally, another BG measure would be taken and a revised treatment calculated, low blood volume means that clinical staff prefer to minimise BG measures.

Statistically, STAR-GRYPHON achieves much better time in the clinically targeted band than the retrospective titration protocols. Although this result stems from a comparison of two patient cohorts of 16 and 25 episodes, respectively, the observed difference between in the mean per-patient percentage time in band for the two group's results is greater than the standard deviation, resulting in a statistical power of over 95%. Further, the median episode

length is 2-3 days, and if the statistical power calculation is repeated on proportions of total BG measurements, this power increases.

Lower difference between groups and higher variability means that the statistical power is lower when retrospective and STAR-NICU results are compared. While the statistical power is lower when median BG is compared, tightness of control has more to do with distribution than a single median value, especially once the median BG is within the target range. While STAR-GRYPHON has much lower %BG<4.0 ($p<0.001$) and %BG<3.0 ($p=0.02$), at least 10 more (at least 26 total) STAR-GRYPHON patients are needed to determine if the lack of patients with a hypoglycaemic episode is statistically significant.

A strength of these clinical results is that performance and safety are defined by more than just average BG measurement and/or hypoglycaemic incidences. However, even the concept of percentage time in band has its limitations. Controversy exists as to the definition of hyper- and hypo- glycaemia, as well as appropriate target ranges (Alsweiler et al., 2007, Cornblath et al., 2000). In addition, differences in glycaemic behaviour and stability can exist within a target range. It has been shown that glycaemic variability may be a predictor for mortality in adult ICU patients (Krinsley, 2008). Further work needs to be done to better and more universally quantify glycaemic control performance and stability.

One of the limitations of this study is that the results are based in a single hospital, the hospital from which the cohort for protocol design was sourced. Other studies have shown that protocol performance in other hospital cohorts can be lower. Currently, STAR-GRYPHON is being used in three other hospitals, two in Auckland, New Zealand and one in Miskolc, Hungary. Future results will give a better indication of its performance across NICU cohorts and clinical practices, particularly as a function of compliance.

The focus of this research is the achievement of tight glycaemic control in this cohort. This study does not attempt to show the effect of the use of insulin therapy to lower BG on outcomes, and would likely be underpowered to do so. If a follow up study on outcomes was carried out, it could perhaps examine outcomes and the tightness of control, but would likely require a much greater cohort.

11.5 Summary

STAR is a model based glycaemic control framework for extremely premature infants. It has successfully been implemented as a standard of care at Christchurch Women's Hospital, New Zealand. The original implementation reduced hyperglycaemia and increased measurements in the targeted range by 16%. Recent implementation of an optimised STAR-GRYPHON protocol has resulted in 76.6% of hourly re-sampled BG measurements in the target band (26% better than retrospective data and 9% better than original STAR NICU protocols), and no incidence of BG<3.5 mmol/L. Comparison with results from the literature is difficult due to the lack of glycaemic control statistics reported, but STAR seems to achieve better or comparable control to published studies with lower incidence of hypoglycaemia. Safer methods will allow further studies to focus on the long term impact on outcome and benefits of glycaemic control on ICU cohorts.

Chapter 12: Cohort Based Insulin Sensitivity Analysis

12.1 Introduction

Quantification of potential future variability in SI enables safe glycaemic control decisions to be made based on a patient's current metabolic state and potential future changes in this state (Chase et al., 2011b, Le Compte et al., 2010b, Le Compte et al., 2013). This variability is clearly evident in Figure 12.1, which shows the hour-hour (S_{In} vs. S_{In+1}) variation in model-based SI obtained from an analysis of clinical data for preterm VLBW and ELBW neonates (Le Compte et al., 2010b). In particular, Figure 12.1 shows a stochastic model of SI, along with an example probability distribution at one S_{In} value, delineating the inter-quartile and 90% ranges.

In fact, the potential variation can be quite large. However, stochastic models are population or cohort based, and thus can be overly conservative for some neonates. Very wide stochastic forecasting bands can thus result in low doses of insulin and persistently high BG. Hence, if sub cohorts of patients with different SI behaviour can be identified, overall stochastic modelling and control can be tightened in an increasingly patient-specific or sub cohort-specific fashion.

This analysis will examine SI distributions between different patient cohorts. The aim is to create stochastic models of SI that not only better account for inter-patient variability over cohorts, but also capture intra-patient variability (per-patient) more accurately. The end-user goal is equally safe, more precise control.

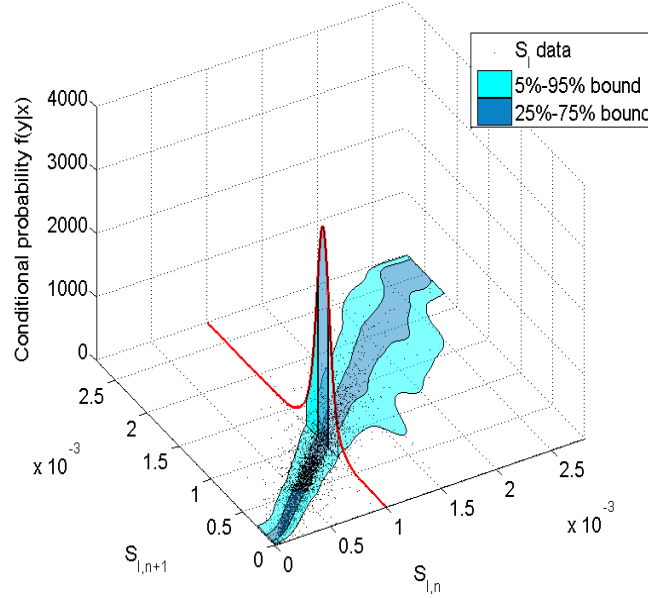


Figure 12.1: Hourly insulin sensitivity variation data with probability bounds and example curve showing probability bounds. Source: (Le Compte et al., 2013)

12.2 Methods

12.2.1 Glycaemic and Stochastic Models

The model used in this analysis is the same as the model presented in Chapter 9, excluding the newly developed gut model. SI profiles are fit using this model, and from these identified SI trajectories stochastic models are created. As the median measurement interval under STAR-GRYPHON is 4 hours, 4 hourly stochastic models are generated using matched SI pairs four hours apart, $\{SI_n, SI_{n+4}\}$.

Stochastic models are constructed using 2D kernel density models applied to a data set of matched SI pairs (Lin, 2007, Lin et al., 2006). As a result, for each discrete SI value within the observed data range, a probability density function is generated. Over the entire SI range, models of 5th and 95th percentile outcomes can be generated. An example stochastic model, generated during this analysis, for 4 hourly predictions using clinical data from 95 patients is shown in Figure 12.2.

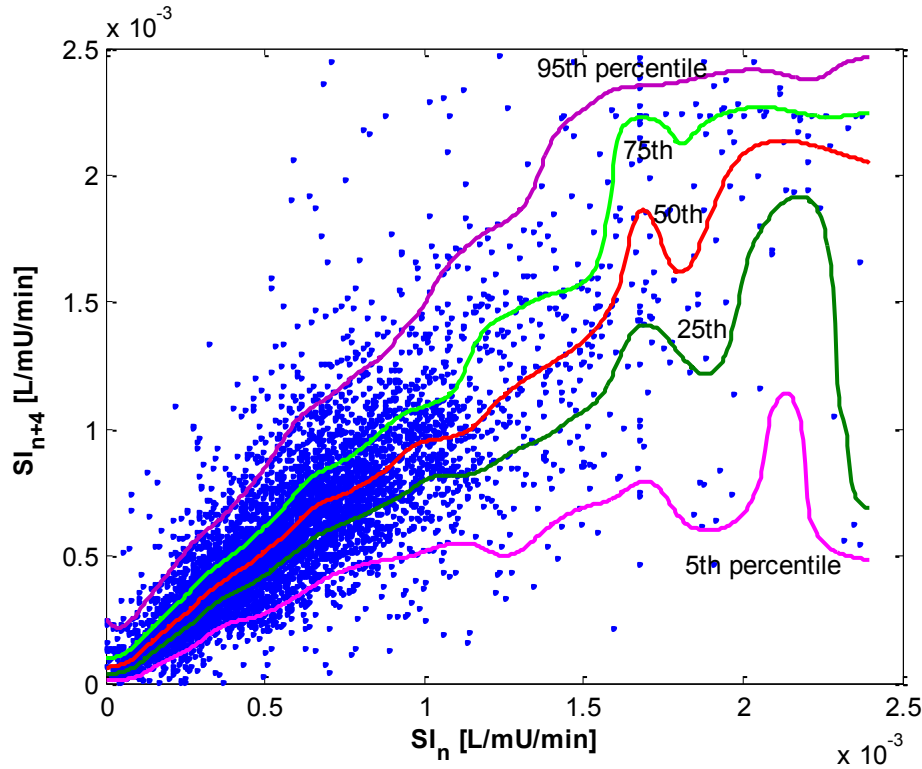


Figure 12.2: Stochastic model for 4 hourly predictions in insulin sensitivity (SI). The model was generated using SI fitted from 95 clinical patients, and shows the 5th, 25th, 50th, 75th, and 95th percentiles for observed changes in SI.

The mathematics of stochastic models are described more fully elsewhere, (Le Compte et al., 2010b, Lin, 2007, Lin et al., 2006), but the kernel density estimator can be modified by a constant c , which effectively adjusts the kernel bandwidth and the degree of smoothing over the data. It can be used to tune stochastic models to more accurately reflect the theoretical 90% coverage of data by the 5th and 95th percentiles, or to smooth model fluctuations in areas of low data density. In this analysis c was set to 1.0.

12.2.2 Clinical data cohorts

SI profiles were fitted from clinical data using Equations 9.1 – 9.13. Three cohorts of glycaemic episodes were compared: 1) retrospective data (N=25); 2) clinical data from control under the STAR-NICU protocol (N=79); and 3) the most recent clinical data cohort treated under the STAR-GRYPHON protocol (N=16). These clinical cohorts were previously presented and discussed in Chapters 10 and 11. Cohort characteristics and control statistics are repeated for clarity in Table 12.1.

Table 12.1: Summary characteristics and glycaemic control statistics for retrospective, STAR-NICU, and STAR-GRYPHON clinical cohorts.

	Retro- spective (R)	STAR- NICU (SN)	STAR- GRYPHON (SG)	P value		
				SN vs. R	SG vs. R	SG vs. SN
n Patients (n male)	22 (7)	40 (22)	13 (8)			
n Episodes	25	54	16			
Total Hours	3098	5014	987			
Number of BG measurements	943	1632	270			
Gestational Age [weeks]	27 [25-27]	26 [25-29]	26 [26-27]	0.82	0.91	0.84
Weight [g]	845 [800-900]	785 [635-950]	908 [820-1030]	0.66	0.24	0.25
Post Natal Age [days]	3 [1-7]	3 [1-12]	3 [2-4]	0.42	0.94	0.45
Measurement interval	3.2 [2.6 - 3.9]	3.3 [2.7 - 3.8]	4.0 [3.8-4.1]	0.98	0.01	<0.001
Insulin rate [IQR] (U/kg/hr)	0.03 [0.02-0.06]	0.04 [0.02-0.08]	0.05 [0.04 - 0.07]	0.06	0.002	0.02
Total glucose delivery rate [IQR] (mg/kg/min)	8.1 [5.7 - 9.7]	7.3 [5.1- 8.5]	7.7 [7.1 - 8.6]	0.50	0.81	0.84
<i>Hourly re-sampled statistics</i>						
Median BG	7.8 [6.6- 9.1]	6.6 [5.5 - 8.2]	6.9 [6.1-7.9]	<0.001	<0.001	<0.001
% BG within 4.0 - 8.0 mmol/L	50.3	67.2	76.6	<0.001	<0.001	0.42
% BG>10 mmol/L	17.7	11.8	5.6	0.01	0.005	0.42
% BG<3.0 mmol/L	0.3	0.7	0	0.28	0.09	0.02

12.2.3 Calculation of Stochastic Model Accuracy

Performance of stochastic models was calculated on a whole cohort and per-patient basis. For each SI value obtained from clinical data, stochastic models were blinded to future SI, and used to generate 5th, 25th, 75th, and 95th percentiles of predicted future SI outcomes. The actual SI outcome was then compared to predicted outcome percentiles, and the percentage of actual SI outcomes that fell within this range was calculated on a per-patient and whole cohort basis. Ideally, a perfect model would capture 90% of each individual patient's variations in the 5-95th percentile interval, as well as the 90% of the variations of the cohort as a whole. Performance can also be assessed by the number of patients whose individual stochastic performance comes close to this ideal.

A cross comparison analysis was carried out with different stochastic models generated from different patient cohorts. Relevance of stochastic models to different clinical cohorts was reflected in the percentage of SI values that fell within predicted 25th-75th and 5th-95th outcome ranges.

12.2.4 Stochastic Forecasting in Prematurity and Gender Sub-Cohorts

Three variables easily identified at the bedside are gender, BW and GA. Cumulative SI distributions are generated and compared for male and female infants, and for sub-cohorts in weight and GA by tertile. Variability in SI is also compared, where variability is reflected in the percentage change in SI from hour to hour, relative to the initial SI value of these matched pairs.

A Kolmogorov–Smirnov (KS) test was used over a full range of possible BW and GA groupings to identify groups with the greatest differences in relative change in SI. This may allow stochastic models to be created based on significant BW or GA groupings.

12.2.5 Statistical analysis

The KS test is used to compare the differences in SI distributions (Matlab[®], kstest2). The Mann-Whitney U test (Matlab[®], ranksum) is used to compare data medians in cohort characteristics or glycaemic statistics. A p-value of 0.05 or less is considered statistically significant.

12.3 Results

12.3.1 Insulin Sensitivity Across Different Cohorts

Table 12.2 shows the percentage of SI within the 5th-95th and 25th-75th percentile prediction ranges for different cohort and stochastic matrix combinations. As expected, when a stochastic matrix is tested against the cohort it was generated from ~ 90% of the predictions fall within the 5-95th percentile range, with differences due to kernel smoothing or regions of lesser data. For these cohort and stochastic model combinations, the percentage of predictions within the 25th-75th range is slightly higher than 50%. If the c value is changed to bring the percentage within the 25th-75th percentile range closer to 50%, then the percentage within the 5th-95th percentile range falls to around 85%.

There is thus a trade-off in percentile distributions when creating the stochastic models. In particular, this trade-off indicates that the distributions are not perfectly matching the kernel density estimator. Since it is the 5th-95th percentile range that drives predictions in the model-based glycaemic control protocols of Chapter 10, stochastic matrices are tailored towards better coverage of the 5-95th percentile range.

When a stochastic model is used with a different cohort from the one it was generated with, the percent of SI in the 5th-95th and 25th-75th percentile prediction ranges is lower, reflecting differences in SI behaviour within and between the three cohorts used. The retrospective and STAR-NICU cohorts are essentially similar, shown by the 86-91% of SI in the 5th-95th percentile prediction range. However, this similarity is not true in the case of the STAR-GRYPHON cohort.

Table 12.2: Whole cohort percentage of SI predictions within the 25th-75th and 5th-95th percentile prediction ranges.

		Stochastic Model population				
		<i>Retro-spective</i> (N=25)	<i>STAR-NICU</i> (N=54)	<i>Retrospective and STAR-NICU</i> (N=79)	<i>STAR-GRYPHON</i> (N=16)	<i>All cohorts</i> (N=95)
Cohort coverage: % time in 5 th -95 th range	<i>Retrospective</i>	90.0	85.4	88.1	69.4	87.3
	<i>STAR-NICU</i>	91.1	87.2	90.0	66.4	89.2
	<i>Retrospective and STAR-NICU</i>	90.7	86.5	89.3	67.6	88.4
	<i>STAR-GRYPHON</i>	96.3	94.9	96.0	89.0	95.4
	<i>All cohorts</i>	91.3	87.4	90.0	69.9	89.2
		Stochastic Model population				
		<i>Retro-spective</i> (N=25)	<i>STAR-NICU</i> (N=54)	<i>Retrospective and STAR-NICU</i> (N=79)	<i>STAR-GRYPHON</i> (N=16)	<i>All cohorts</i> N=(95)
Cohort coverage: % time in 25 th -75 th range	<i>Retrospective</i>	52.8	46.4	48.8	33.0	46.6
	<i>STAR-NICU</i>	55.0	51.6	53.3	35.8	51.8
	<i>Retrospective and STAR-NICU</i>	54.1	50.0	51.6	34.7	49.8
	<i>STAR-GRYPHON</i>	68.4	64.4	64.4	53.5	61.6
	<i>All cohorts</i>	55.7	51.2	53.0	36.7	51.1

In all cases, the STAR-GRYPHON derived stochastic model resulted in much narrower prediction ranges when used with other cohorts, resulting in much lower percent time in both the 25th-75th and 5th-95th percentile ranges. On the other hand, when stochastic models derived from other data cohorts was used with the STAR-GRYPHON cohort, the percentage of predictions within the 25th-75th and 5th-95th percentile ranges was much higher than 50% and 90% respectively. These results indicate that other stochastic models are overly conservative for this cohort, despite no statistically significant differences in Table 12.1 in GA, BW, or PNA.

Figure 12.3 shows the cumulative distributions of SI for all data cohorts. All data cohorts significantly differ from each other in whole cohort SI distribution ($p < 0.001$). STAR-GRYPHON and STAR-NICU patients have lower ($p < 0.001$) and less variable ($p < 0.001$) SI than retrospective data, and the STAR-GRYPHON cohort is the least variable ($p < 0.001$).

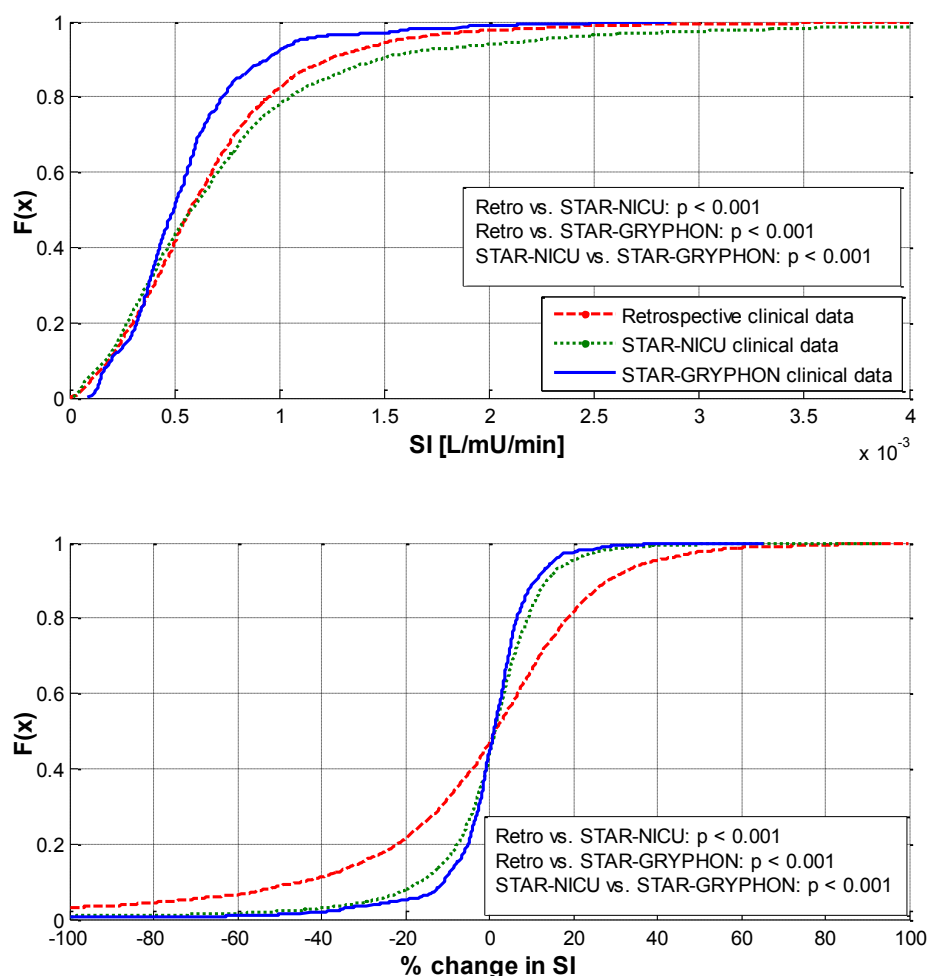


Figure 12.3: SI distribution and variability for Retrospective, STAR-NICU, and STAR-GRYPHON clinical data cohorts. The legend applies to both plots.

12.3.2 Per-patient Stochastic Model Performance

Figure 12.4 shows the per-patient performance of a whole cohort (N=95) stochastic model used across the entire patient cohort (N=95). This model had a whole cohort percentage time in the 5th-95th percentile band of 89.3%, indicating the stochastic model adequately reflected cohort behaviour as a whole. Of this cohort, 50 (53%) of patients had 90% or greater per-patient percentage time in the 5th-95th percentile band. In addition, 78 (82%) of patients had 80% or greater per-patient percentage time in the 5th-95th percentile band, while 1 patient had less than 50% of per-patient percentage time in the 5th-95th percentile band. These results indicate that per-patient SI evolution is highly patient specific, and while stochastic models adequately capture whole cohort behaviour, a stochastic model is not itself necessarily reflective of any single patient's SI behaviour.

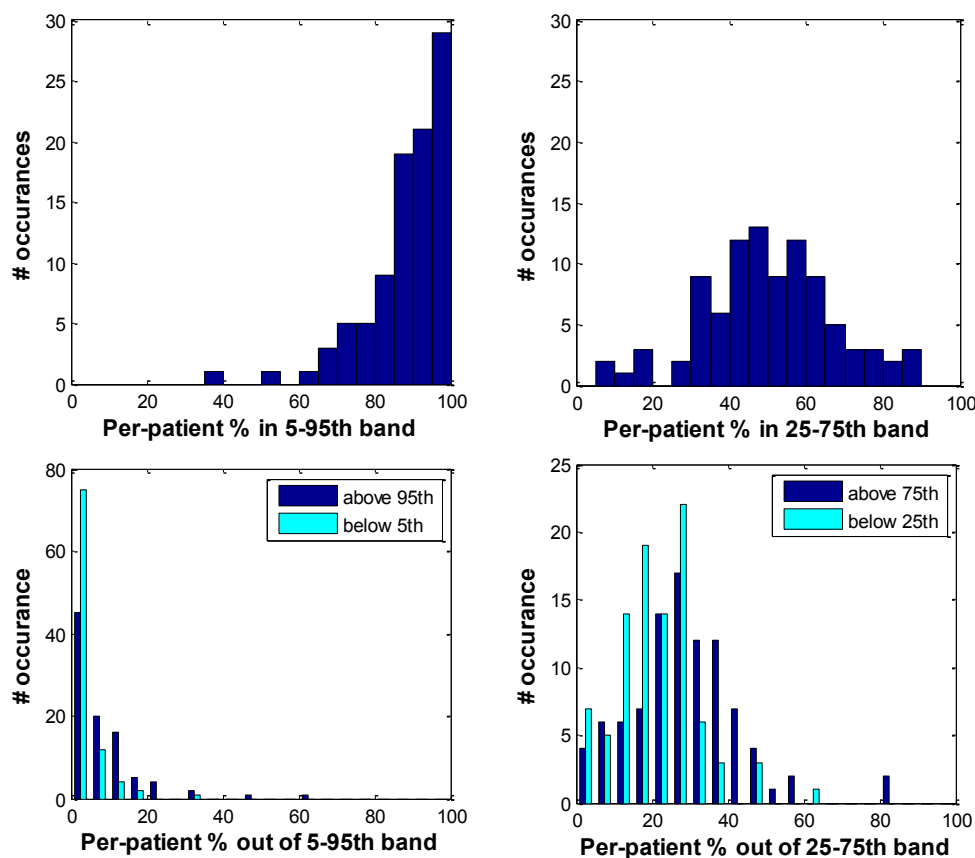


Figure 12.4: Per-patient performance across the entire patient cohort (N=95) of stochastic models generated from the entire patient cohort (N=95).

Figure 12.5 is similar to Figure 12.4, but reflects SI behaviour within the retrospective cohort only. Again, 53% (13 of 25 patients) had 90% or more SI changes within the predicted 5th-95th percentile range. In addition, 21 (84%) of patients had 80% or more SI changes within the predicted 5th-95th percentile range. This result reflects how even within more similar clinical sub cohorts there is significant inter-patient variability. Thus, the individuals create a stochastic model, but for this cohort, the model is not fully patient-specific.

Figure 12.6 shows per-patient performance for the STAR-GRYPHON cohort when a stochastic model generated from STAR-NICU and retrospective data is used. This combination was one that showed a clearly over-conservative stochastic model, with 96% of whole cohort SI within the 5-95th percentile prediction range. 62% (10 of 16) patients have 95% or more of SI within the 5-95th percentile prediction range, and 81% (13 of 16) have 80% or more SI in the 5-95th percentile prediction range. However, despite this high percentage of SI within the target range for most patients, 1 patient still was still under-represented, with 60-65% of their SI within the 5th-95th percentile prediction range.

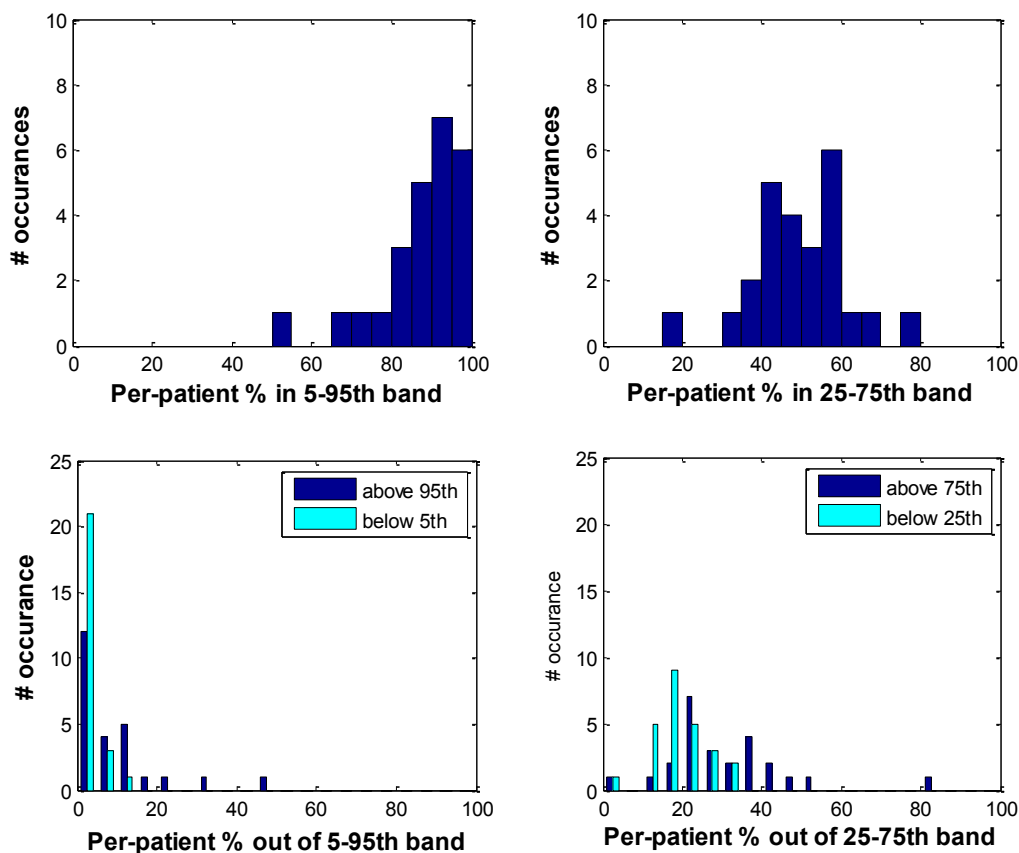


Figure 12.5: Per-patient performance across the retrospective patient cohort (N=25) of stochastic models generated from the retrospective patient cohort (N=25).

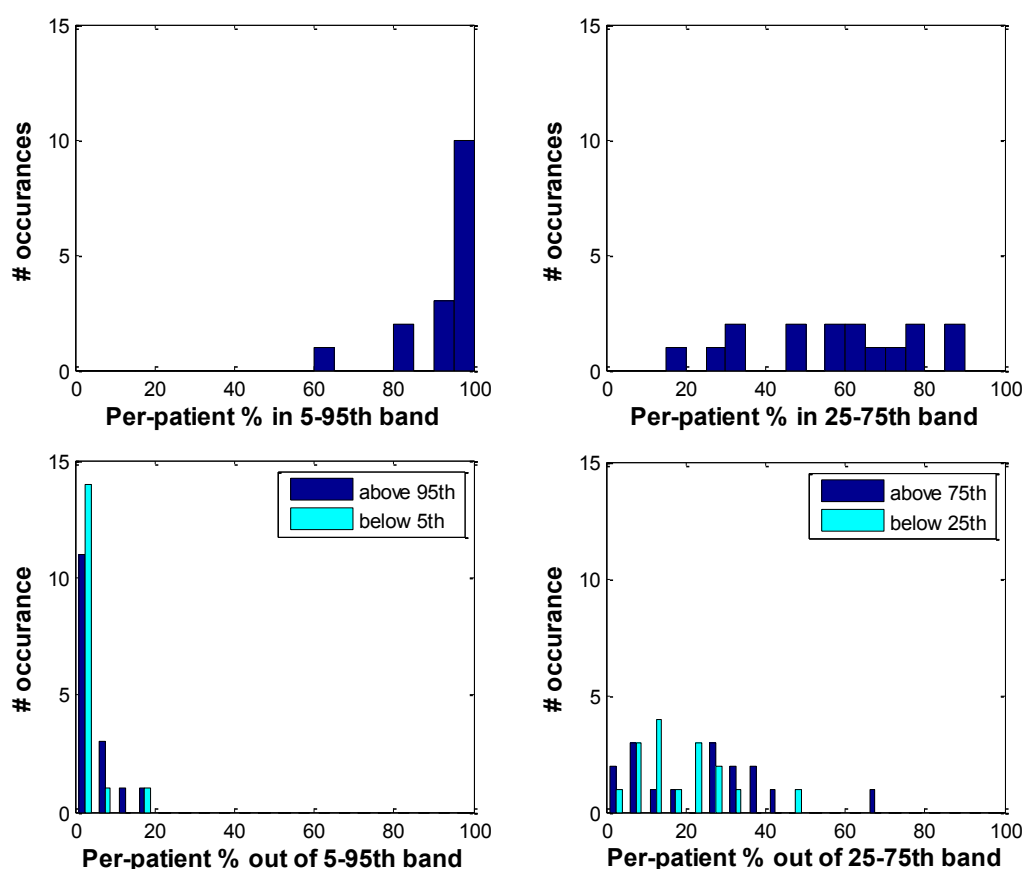


Figure 12.6: Per-patient performance across the STAR-GRYPHON patient cohort (N=16) of stochastic models generated from the STAR-NICU and retrospective patient cohorts (N=79)

Patients with more than 5% of their SI above the 95th percentile prediction, or more than 5% of their SI below the 5th percentile prediction, were examined, and no trends in BW, weight, GA, or PNA were found. Thus, SI evolution is highly patient-specific, and dependent on more factors than examined in this analysis, and is not dependent on easily measured admission or patient demographics.

12.3.3 Stochastic forecasting and SGA/AGA cohorts

Of 79 total patients, 14 were SGA. Figure 12.7 shows cumulative distributions in SI and variation in SI by SGA and AGA cohorts. SGA infants have higher insulin and less variable SI ($p < 0.001$). This outcome is surprising, as SGA infants are commonly regarded as sicker than their AGA counterparts, and thus one would expect greater stress response and greater dysfunction due to prematurity that would lead to greater variability.

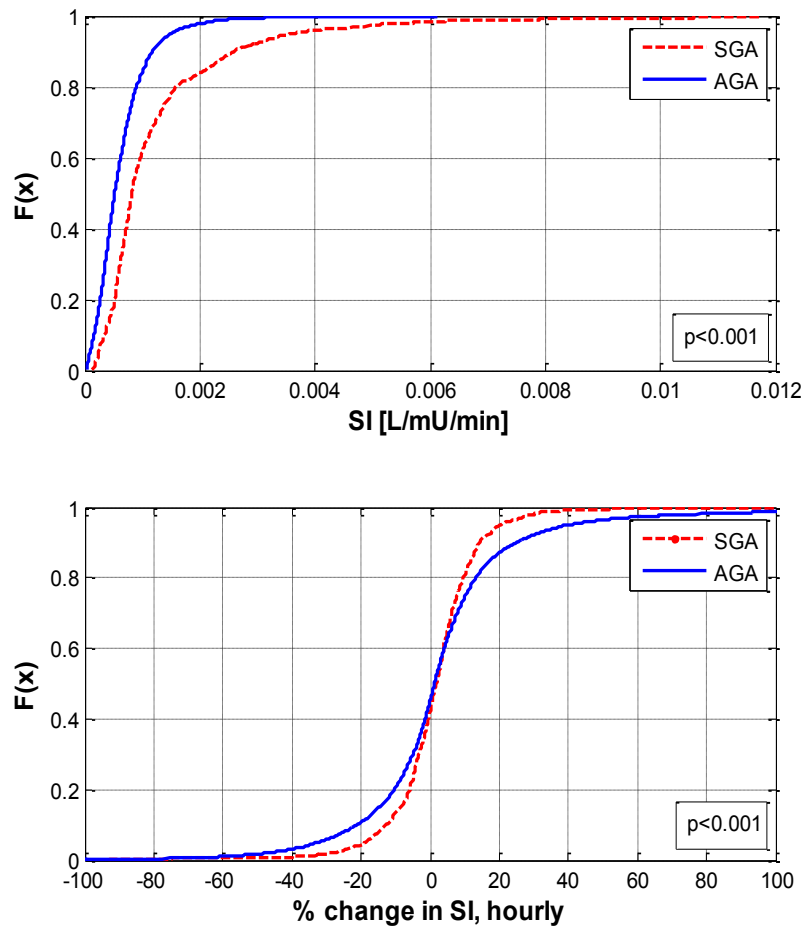


Figure 12.7: Cumulative distributions of insulin sensitivity (SI) and variation in insulin sensitivity on the basis of weight for gestational age. The SGA cohort is small for gestational age, defined as BW below the 10th percentile of weight for their gestational age. AGA is the appropriate for gestational age cohort, and consists of all infants of birth weight greater than the 10th percentile for their gestational age. Birth weight percentiles were determined from (Beeby et al., 1996).

12.3.4 Gestational Age and Weight Sub-Cohorts

The entire data population was divided into sub-cohorts based on the population tertiles in weight and gestational age (767g and 920g, and 25.4 wk and 27.0 wk, respectively). Figure 12.8 shows cumulative distributions in SI and SI variability for patient weight and GA. SI is lower with increasing prematurity and lower birth weight. Differences in variability between cohorts are more distinct when cohorts are divided by GA, but with no distinct trend in GA and variability. Table 12.3 shows that tertiles have relatively consistent distribution of patient numbers and SI hours between tertiles, eliminating bias due to low numbers.

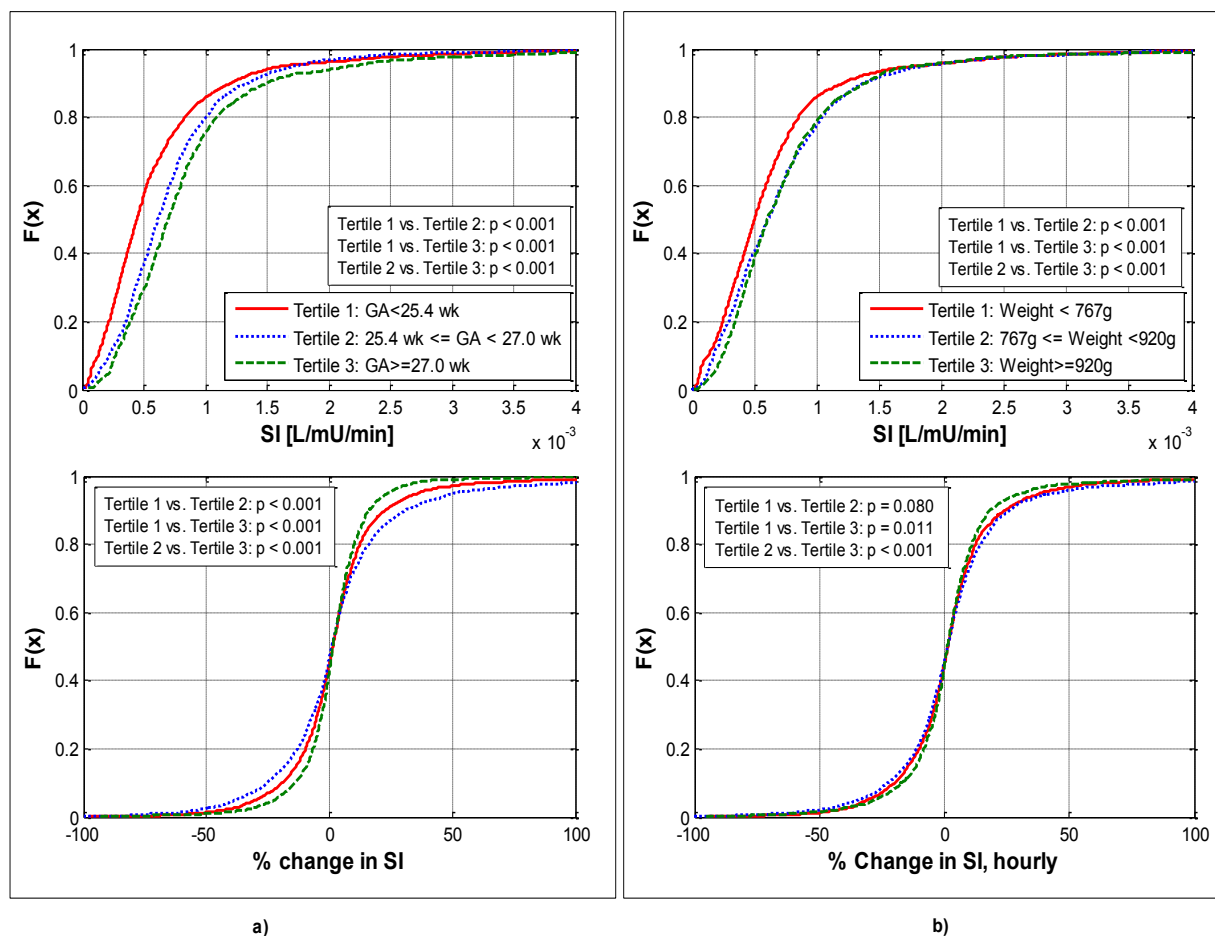


Figure 12.8: Insulin sensitivity (SI) and insulin sensitivity variability with tertiles in a) gestational age (GA), and b) weight.

Table 12.3: Patient numbers and SI hours for tertiles of gestational age (GA) and weight

<i>Weight Tertiles</i>			
		Number hours data	Number of patients
Tertile 1	Weight < 767g	3454	79*
Tertile 2	767g \leq Weight < 920g	2766	64*
Tertile 3	Weight \geq 920g	2980	85*
	Total	9200	95
<i>Gestational Age Tertiles</i>			
Tertile 1	GA < 25.4wk	3838	31
Tertile 2	25.4wk \leq GA < 27.0wk	2871	31
Tertile 3	Weight \geq 27.0wk	2491	33
	Total	9200	95

* Most patients had more than 1 weight measurement

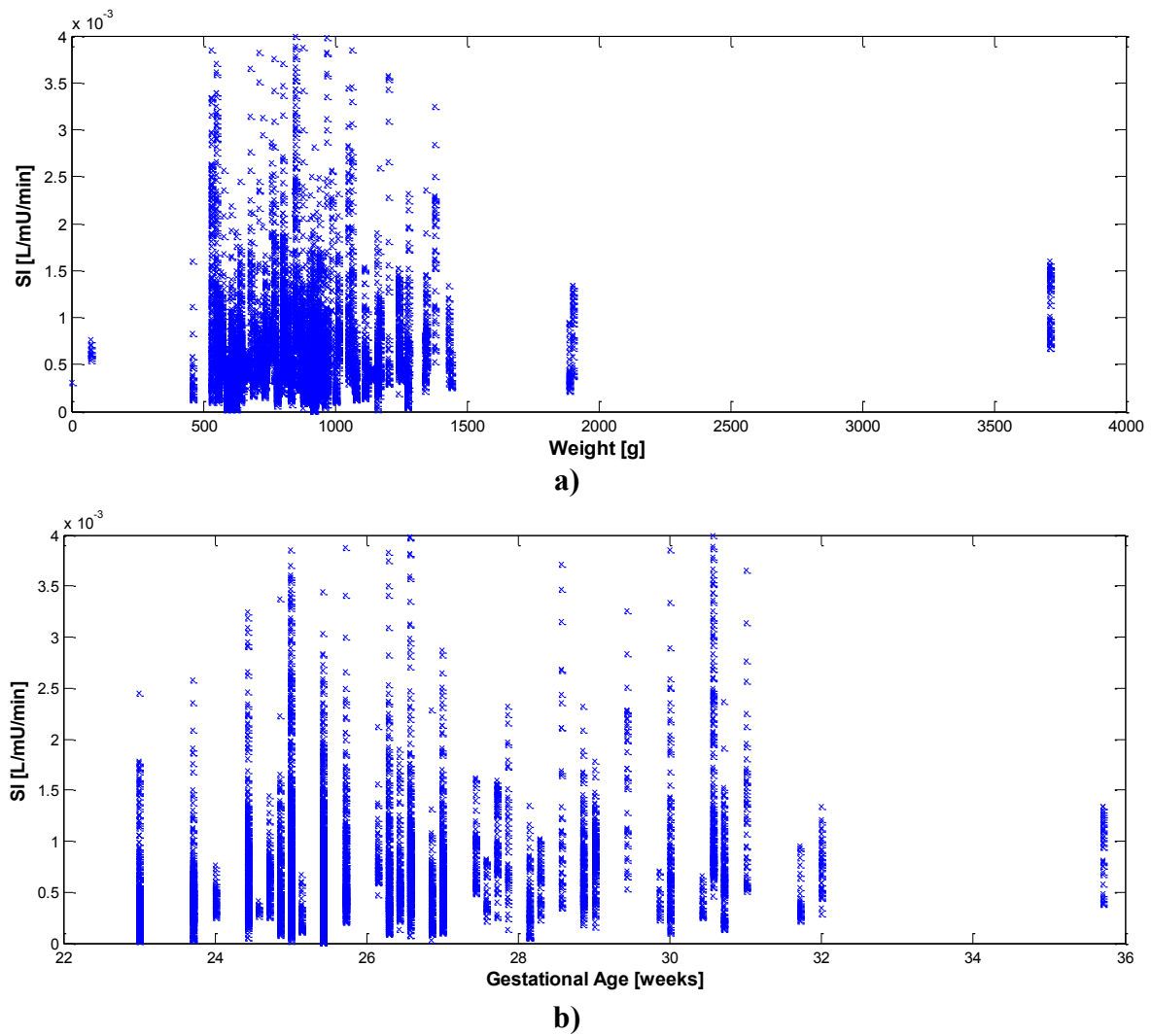


Figure 12.9: Insulin Sensitivity with a) patient weight and b) gestational age.

SI is highly variable with weight, as shown in Figure 12.9. The data cohort was subdivided into two cohorts by a cut off weight that was made to vary between 600 and 1300g in 5g increments. It was found that almost all resulting SI cohorts across the weight range were statistically different in absolute SI ($p \leq 0.01$). Thus, weight cannot be used as a basis for creating different stochastic models for different cohorts within the dataset. However, SI variability is statistically different in infants of weight $< 650\text{g}$ and weight $> 805\text{g}$. If SI variability was used instead of absolute SI in prediction, weight divisions could be used to define sub cohorts in SI behaviour.

The same result of high variability was true for GA. The cohort was divided into two sub-cohorts by GA, with the cut off weight made to vary between 24.0 and 30 weeks in steps of 0.5 weeks. All SI sub cohorts were statistically different ($p < 0.001$). However, SI variability is

statistically different in infants of $GA < 25.5$ weeks and $GA > 26.5$ weeks. Thus, if SI variability was used instead of absolute SI in prediction, GA divisions can be used to define sub cohorts in SI behaviour.

12.3.5 Insulin Sensitivity and Sex

Figure 12.10 shows a difference in SI distributions between the sexes ($p < 0.001$), with female infants showing lower model-based SI, and higher variability. This difference holds even if the non-sex differentiated model for insulin secretion of Equation 4.9 is used ($p < 0.001$). The sexes are not different in GA, PNA, or BW ($p \geq 0.55$), as shown in Table 12.4. However, they are different in the quality of their control. The male infants had lower median BG ($p < 0.001$), less hyperglycaemia ($p = 0.03$), and higher time in band ($p = 0.06$). Thus, the female infants were more insulin resistant and had poorer glycaemic control, likely reflecting greater sickness and injury than the male infants.

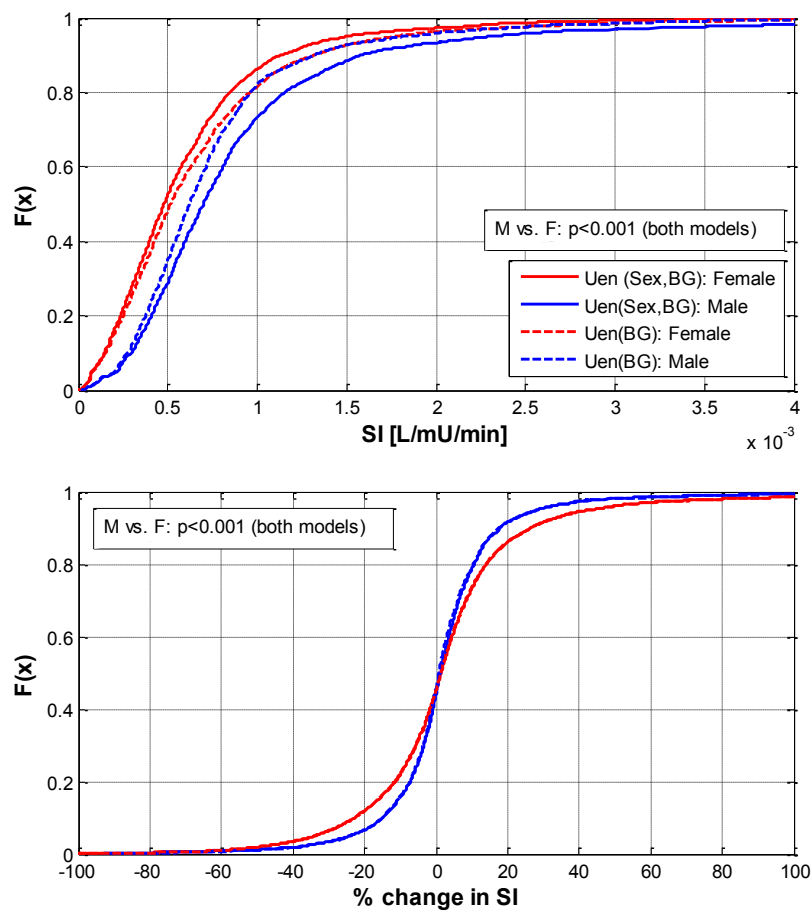


Figure 12.10: Insulin sensitivity (SI) and glycaemic variability between male and female infants. To ensure differences were not directly model-induced, SI was also fit using the non-sex differentiated model of Equation 4.9.

Table 12.4: Characteristics and glycaemic control statistics for male and female cohorts

	Female	Male	p
N	51	44	
Gestational Age (GA) [weeks]	24.4 [25 – 27.4]	25.8 [25.0 – 27.8]	0.97
Post Natal Age (PNA) [days]	4 [2 – 8]	4 [2 – 7]	0.55
Birth Weight [g]	825 [635 – 933]	830 [635 – 940]	0.93
<i>Clinical glycaemic control stats:</i>			
Median insulin rate [IQR] [U/kg/hr]:	0.04 [0.02 - 0.08]	0.040 [0.02 - 0.07]	0.72
Median glucose rate [IQR] (mg/kg/min):	7.3 [4.6 - 8.9]	7.9 [6.3 - 9.2]	0.07
Median BG [mmol/L]	7.3 [5.9 - 8.9]	6.7 [5.8 - 8.2]	<0.001
% BG within 4.0 - 8.0 mmol/L	59.8	69.7	0.06
% BG>10 mmol/L	14.7	8.6	0.03
% BG<4.0 mmol/L	2.9	2.6	0.21

12.4 Discussion

12.4.1 Clinical Cohorts in Stochastic Models

Comparison of per-patient percentile band coverage for different cohort based stochastic matrices has shown that the cohorts are effectively captured by similar stochastic models, despite differences in SI distributions ($p<0.001$). The addition of 54 patient episodes (N=25 to N=79) improved per-patient and whole cohort stochastic model coverage in the STAR-NICU and retrospective cohorts slightly, but not to an extent that is likely to be clinically significant in use. This similarity in coverage with the use of different cohort stochastic forecasting verifies that the original data set was sufficiently representative of the STAR-NICU population as previously thought, where initial work with stochastic matrices in adult ICU patients indicated N = 25 would be suitable (Lin et al., 2006).

The high proportion of coverage above 90%, and in particular above 95% in the 5-95th band when using the STAR-NICU and retrospective data derived stochastic models on the STAR-GRYPHON cohort suggests that this cohort has different SI behaviour than the previous STAR-NICU cohort. This outcome was also seen when SI distributions were compared between cohorts, with the STAR-GRYPHON cohort displaying higher SI and lower SI variability than STAR-NICU and retrospective cohorts ($p<0.001$). This behaviour may result

from underlying differences in patient condition or patient demographics. Alternatively, it may be a result of greater consistency in measurement interval and fewer changes in nutrition reducing noise in the SI profile. Both causes are likely true.

Previous discussion in Chapter 11 identified lower incidence of hyper-and hypo-glycaemic extremes, revealing slightly over-conservative stochastic models. Until the STAR-GRYPHON cohort is larger, reaching approximately 25 patients (Lin et al., 2006), the STAR-NICU and retrospective data based stochastic cohorts are adequate for glycaemic control. In addition, they seem to err on the side of safety through over-conservative SI predictions, resulting in more than 90% of SI within the 5th-95th percentile prediction range.

Per-patient percent time in the 5th-95th percentile range showed that patient evolution of SI is highly patient specific. The high proportion of coverage above 90%, and in particular above 95% in the 5-95th band suggests that this band width is determined by the behaviour of a relative few patients. Ideally, for patient specificity, the majority of per-patient coverage would be close to the target 90%. The patient coverage of the 25-75th percentiles is much wider, indicating significant inter-patient variability and lack of patient-specific coverage within these central tendency bounds. Overall it seems that inter-patient variation is more significant than intra-patient variation as a limiting factor in this stochastic forecasting model, and that a relative few more variable patients are quite different in behaviour.

Population based stochastic models are clearly limited by their inability to adequately capture patient-specific SI behaviour. However, their use has proven safe in clinical usage, with additional constraints around insulin dosage rates limiting the impact of SI values outside the patient range. More patient-specific prediction methods, such as auto-regressive based methods, have been examined elsewhere (Le Compte, 2009), but suffer from the significant drawback that rapid changes in SI can render such data based models obsolete. In addition, much narrower prediction ranges than stochastic models provide, more readily results in more aggressive insulin dosing that can lead to hypoglycaemia when patient condition changes rapidly. Future work should examine other more patient specific forecasting methods that can ameliorate these risks.

12.4.2 Insulin Sensitivity and Patient Bedside Metrics

Bedside metrics offer consistent, objective classification. SI was highly variable across all BW and GA ranges. Further, for patients not well represented by stochastic models in the per-patient percentage coverage statistics, there was no consistent trend with GA or BW to characterise these patients.

However, SI was found to be higher and less variable in SGA infants, a result also seen in Chapter 7 with HINT patient data. This result is unusual in that SGA infants are generally considered to be more at risk, with more higher morbidity and worsened outcomes than their AGA counterparts (Girsén et al., 2015, Hei et al., 2014). This result may reflect the relatively small number of SGA infants (14 of 79), and warrants further investigation with a larger patient cohort. If indeed SGA infants have different SI behaviour than their AGA counterparts, then stochastic models can be created to capture this behaviour of an easily and objectively identified cohort.

SI was found to differ between the sexes. Female infants were found to have lower SI and higher SI variability than their male counterparts, also reflected in the clinical glycaemic control statistics, with lower BG within the 4.0-8.0 mmol/L target range and higher incidence of hyperglycaemia ($\%BG < 10.0$ mmol/L.). This result is counter to expectation, as premature male infants are generally regarded as sicker than premature female infants, and with worsened outcomes such as higher mortality (Stevenson et al., 2000), and poorer neuro-developmental outcome (Henderson-Smart et al., 2006). Further analysis is required to distinguish whether this is a consistent trend in this NICU population, or simply a result of unusually high numbers of sicker premature infants.

12.5 Summary

Analysis of model-based insulin sensitivity distributions for different clinical cohorts revealed that retrospective, STAR-NICU, and STAR-GRYPHON cohorts all differ in their SI behaviour ($p < 0.001$). However, retrospective and STAR-NICU cohorts are both effectively captured by a stochastic model based on their combined SI behaviour. The STAR-GRYPHON cohort to date has higher SI and lower variability than STAR-NICU and retrospective cohorts ($p < 0.001$). This outcome may result from more consistent measurement intervals and lower variability in feed rates, or from differences in the underlying clinical characteristics of the patients not evident in the fundamental demographic data available.

Both reasons are likely true. However, more STAR-GRYPHON patients are needed to generate effective stochastic matrices for this population alone. In addition, though slightly over-conservative, the STAR-NICU and retrospective data based stochastic matrix currently in use is adequate and safe in control.

Model-based SI was found to be higher in SGA and male infants. These results are surprising, given that these two cohorts usually are associated with greater sickness and injury, and worsened outcomes. Further analysis is required to determine whether they are typical of their NICU sub-cohort, or the result of unusually high numbers of healthier SGA or male infants. Model-based SI was found to be highly varied with birth weight and gestational age, resulting in no tangible benefit from stochastic models for weight or gestational age based clinical sub-cohorts.

Chapter 13: Conclusions

13.1 Major Chapter Outcomes

In neonatal intensive care hyperglycaemia is a common complication of prematurity, affecting up to 70% of very premature infants. Hyperglycaemia has been associated with mortality and worsened outcomes in neonatal intensive care. However, it is still debated whether hyperglycaemia is a reflection of worsened patient condition or a cause in itself, with both likely true.

Studies examining glycaemic control outcomes in this cohort are relatively rare, and plagued by increased incidence of hypoglycaemia, which is also associated with worsened outcomes. The fragility of the cohort further limits data, studies, and the ability to learn from the results. Current clinical practice regarding glycaemic control differs between units, and insulin therapy and glucose restrictions are two approaches used to treat hyperglycaemia.

Insulin therapy has been well established as an effective treatment for hyperglycaemia that also allows for increased glucose tolerance and higher weight gain. Clinical insulin therapy protocols are typically ad hoc and/or titrate insulin with blood glucose level. These approaches fail to adequately account for differences between patients and changing patient condition over time, resulting in poor control and increased risk of hypoglycaemia.

Model-based control uses a time varying insulin sensitivity parameter to describe changing patient response to insulin over time, and between patients. Only one such model-based approach exists in premature neonatal cohorts. The object of this thesis was to improve on this approach by more comprehensive modelling of patient-specific aspects of glucose-insulin physiology, and to develop and optimise a STAR glycaemic control protocol for neonatal intensive care.

The NICING (Neonatal Intensive Care Insulin-Nutrition-Glucose) model was developed from a similar fundamental model used in adult intensive care. In particular the modelling of insulin, glucose, and the breakdown and absorption of enteral glucose was revisited from the prior NICU model and adult models. Models were made specific to the very/extremely premature neonatal intensive care unit cohort using relatively rare clinical data and extensive analysis of relevant results in the literature.

Insulin secretion in this cohort was analysed using C-peptide measurements. This approach is extremely novel in this cohort, as previously only absolute insulin concentrations and insulin AUC estimates have been used to indirectly assess pancreatic function. Insulin secretion was found to be highly variable across the range of subjects and clinical variables. However, insulin secretion was found to be twice as high in female than male infants, and was also found to increase with increasing blood glucose concentration. Simple, but representative, first of their kind, linear models of insulin secretion as a function of blood glucose were created for male and female very premature infants.

Insulin clearance is another unknown that increases apparent variability in response to treatment. Separate insulin clearance pathways were modelled using C-peptide and insulin concentrations in very premature infants. In particular, clearance by the liver and kidneys (n_L and n_K respectively) was examined, as well as cellular degradation of insulin (n_C), and diffusion of insulin between plasma and peripheral fluid compartments (n_I). Renal clearance of insulin was modelled using the literature surrounding kidney function in this cohort. Based on age specific changes in kidney function, this renal clearance of insulin is also allowed to change with patient birth weight and post-natal age. Improved liver clearance, cellular degradation, and trans-capillary diffusion of insulin were then determined using the NICING model structure and C-peptide and insulin concentration measurements from a cohort of very premature infants.

As part of the insulin clearance modelling, compartment fluid volumes were analysed. While blood plasma volume was found to remain relatively constant, the extracellular fluid compartment was found to contract significantly in the days following birth. Modelling this effect enables the wider insulin kinetic model to be modified to better reflect patient age.

Within glucose modelling two key aspects of glucose modelling were examined: 1) endogenous glucose production (EGP); and 2) non-insulin mediated glucose uptake, predominantly by the central nervous system (CNS). EGP was examined using a meta-analysis of data from the literature. EGP was found to be highly variable across a number of patient metrics. EGP was found to be suppressed with increasing glucose infusion, and in most cases be suppressed at higher blood glucose concentration. However, after further fitting and control performance based analysis, it was found that for the purposes of glycaemic control, EGP is best modelled as a population constant ($EGP = 6.0 \text{ mg/kg/min}$). This result reflects the high variability in observed EGP rates in the literature. It also reflects the fact that

these rates were measured in non-hyperglycaemic, otherwise healthy, very premature infants, which may confound the issue.

Non-insulin mediated CNS uptake was examined indirectly through the modelling of brain mass in very premature infants. Two models were compared. First brain mass was treated as a simple 14% of body weight, and second, brain mass was modelled as a function of head circumference. It was found that using head circumference to estimate brain mass generally resulted in larger brain mass estimations than when it was estimated as a proportion of body mass (median 121g vs. 112g). This outcome was especially true in small for gestational age (SGA) infants. However, there was no control benefit to be gained from using head circumference over body mass to estimate brain mass, irrespective of whether the infant was SGA or not. As such, brain mass modelled as 14% of body mass is adequate for control, and, unlike head circumference based models, does not increase clinical workload or complexity.

A two compartment model of glucose transfer through the stomach and absorption in the gut was implemented. Two key parameters are determined, the rate constant for gastric emptying from the stomach, d_1 , and the rate constant for glucose absorption from the gut to the bloodstream, d_2 . Gastric emptying half lives are reported for a range of different gestational ages in the literature, with slower gastric emptying in more premature infants.

Using CGM data from preterm and term infants, the rate constant for glucose absorption from the gut to the blood stream was examined. In this analysis the rate constant $d_1 = 0.035 \text{ min}^{-1}$, which corresponds to a gastric emptying half life of 20 minutes, was used. From glucose appearance in CGM data, the rate constant for glucose absorption was determined to be $d_2 = 0.014 \text{ min}^{-1}$, corresponding to an absorption half life of 50 minutes. This absorption rate constant was not found to change with gestational age. Though determined from premature and term infants, this value is a better estimation of intestinal glucose absorption in very/extremely premature infants than adult values used previously. In very/extremely premature infants the gastric emptying rate constant can be set to the slower $d_1 = 0.017 \text{ min}^{-1}$ (40 minutes half life) to better reflect their condition.

Following model development virtual patients were created using the new NICING model fitted to clinical data. A virtual patient consists of a model based SI trace, and was used in virtual trials. Variations on a STAR (Stochastic Targeted) protocol were tested in virtual trials. These STAR protocol variations utilised stochastic forecasting models for insulin

sensitivity to generate percentiles of likely blood glucose outcomes. Insulin doses were chosen such that predicted blood glucose outcome percentiles met certain criteria. The chosen protocol placed the 5th percentile of likely blood glucose outcomes within a 4.0-4.4 mmol/L target range. Adaptive limits in the limit on the increase in insulin infusion rate were applied with blood glucose concentration. The final protocol is known as the STAR-GRYPHON protocol, and clinically validated virtual trials showed a performance of 76.4% hourly re-sampled blood glucose measurements within the clinically targeted 4.0-8.0 mmol/L range, and low incidence of hypoglycaemia (0.1% BG<2.6 mmol/l with 2 of 79 (2.5%) virtual patients with at least one re-sampled BG<2.6 mmol/L). .

The STAR-GRYPHON protocol was implemented as a customised tablet computer-based application in Christchurch Women's Hospital NICU from January 2013 to January 2015. A total of 13 patients, totally 16 glycaemic episodes, were treated using STAR-GRYPHON during this time. Percentage time in band (re-sampled hourly) was 76.6%, with an average measurement interval of 4 hours. There were no instances of blood glucose measurements falling below 3.5 mmol/L, and no incidences of severe hypoglycaemia. Performance of STAR-GRYPHON improves on retrospective clinical percentage time in band by 26%, and is very near to pre-trial virtual trial results. Thus, STAR-GRYPHON is thus safer and more effective at providing glycaemic control than previous implementations and clinical practice.

Using this clinically validated model, insulin sensitivity distributions were found to differ between the retrospective, STAR-NICU, and STAR-GRYPHON cohorts. Retrospective and STAR-NICU cohorts were found to be adequately represented by a stochastic model based on retrospective and STAR-NICU data. However, this stochastic model was found to be overly conservative for the STAR-GRYPHON cohort, with 96% of whole cohort insulin sensitivity within the 5th-95th percentile prediction range. Further, 62% of patients had a per-patient percentage of SI within this range greater than 95%. This result may reflect underlying differences in the STAR-GRYPHON cohort compared to previous cohorts, or be a result of more consistent measurement interval and lower variability in nutrition delivery. Both are likely true. Approximately 10 more patients are required to generate a STAR-GRYPHON specific stochastic model for clinical use. In the meantime, the current retrospective and STAR-NICU data based stochastic model is safe and adequate for clinical use.

13.2 General Conclusions

At the outset, this thesis aimed to take an existing model-based glycaemic control framework used in neonatal intensive care, and improve on its performance through more comprehensive modelling of glucose-insulin dynamics and better quantification of sources of variability. This goal was achieved using a novel clinical data set and detailed literature analysis. As a result, the NICING model has been developed, which, in particular, has more comprehensive and detailed modelling of insulin secretion and clearances. The NICING model tightens control, indicating that sources of insulin sensitivity variability have been better captured and accounted for in the model, providing a much better representation for accurate, safe, insulin dosing.

A model-based protocol for glycaemic control in neonatal intensive care was developed, namely the STAR-GRYPHON protocol. Clinical results show significant improvement over retrospective data and previous protocol implementations, with no incidence of severe hypoglycaemia, a result unmatched in the literature to date.

The majority of the literature surrounding glycaemic control in premature infants to date has focused on clinical outcomes. The weakness of this approach is that development and protocols for first achieving control have been overlooked. As a result, outcomes have been confounded by poor control and increased incidence of hypoglycaemia. The STAR framework as a glycaemic control tool has been previously shown to be safe and effective in adult and neonatal intensive care, and STAR-GRYPHON further improves on all glycaemic control in the literature to date. It can thus be used in a wider clinical and research context to assess the long and short term effects of glycaemic control on clinical outcomes such as morbidity, length of hospital stay, and length of mechanical ventilation.

A major conclusion of this work is that the glucose-insulin system in preterm infants is very complex, with many immeasurable factors influencing it. This result is informally expressed in Figure 13.1 (Dickson et al., 2014), where the scatter in clinically measured data meant that more traditional models were often hard to fit. One of the temptations when faced with such complex systems is to increase model complexity to capture every potential influence. However, this thesis demonstrates that relatively simple models can be clinically applicable and accurate. The very simplicity of this model means that it can be used to significantly improve the standard of care without significantly increasing clinical workload, or demanding more measurements than are ethical or practically feasible.

Finally, this thesis lays out a model-based approach to dosing insulin in neonatal intensive care for the purposes of glycaemic control. In neonatal intensive care in general, drug therapy is often *ad hoc*, using dosage regimes extrapolated from paediatric or adult intensive care (Allegaert and van den Anker, 2014). This thesis demonstrates that using known physiological pathways to generate models can be an effective and relatively non-invasive way to safely and effectively improve drug therapy in this fragile cohort.

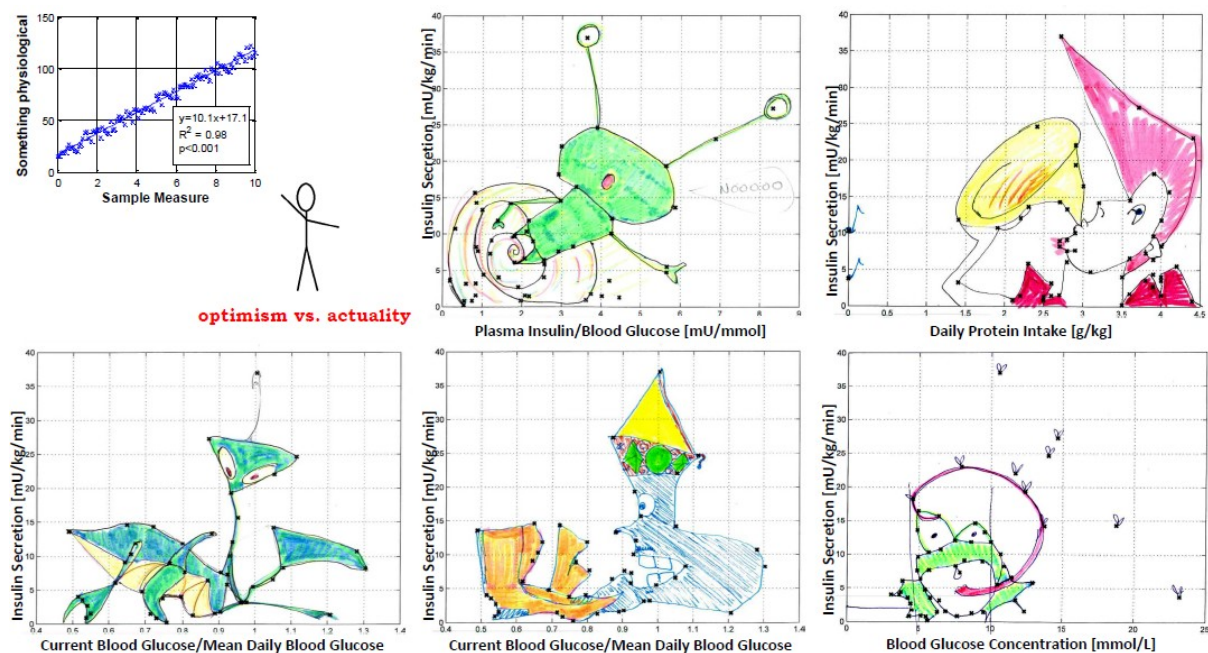


Figure 13.1: Humans are horribly variable. Human physiology is complex and pathways rarely act in isolation. As such, clinical data is often highly variable in nature, with outcomes dependent on a number of different clinical and patient specific aspects, such as when comparing insulin secretion to a number of physiological markers. Figure source: Dickson et al. (2014)

Chapter 14: Current and Future Work

A glycaemic control framework has been developed for neonatal intensive care, and implemented in Christchurch Women's Hospital. Initial results show improved care over retrospective data and results from the literature. Further avenues of exploration have been opened up by the work in this thesis.

14.1 Use in Different NICU Centres

STAR-GRYPHON is proving effective in the Christchurch Women's Hospital Neonatal Intensive Care Unit, and is currently used as a standard of care. However, for further proof of its efficacy, its use must be expanded into other NICU's, both in New Zealand, and overseas.

Within New Zealand, STAR-GRYPHON is currently being used as the glycaemic control tool behind the tight glycaemic control arm of the HINT2 trial, a follow up trial to the published HINT trial. This trial is taking place in Auckland NICU's, and will attempt to assess the effect of tight glycaemic control on outcomes in premature infants. Clinical enrolment in the trial began in July 2015. However, to date, only one infant has been randomised to the tight glycaemic control arm of the trial. Future work will require a long term follow up on the performance of STAR-GRYPHON within this different clinical context.

Outside of New Zealand, STAR-GRYPHON is being trialled in Miskolc County Hospital NICU, Hungary. To date, staff in compliance has resulted in only two 12 hour periods of babies on insulin under the STAR-GRYPHON protocol. In both cases blood glucose was brought within the band with no incidence of hypoglycaemia, before a nursing shift change meant that STAR-GRYPHON was no longer used. Future work will require further nurse training and to improve compliance and assess performance in a clinical context that is potentially very different from that in New Zealand.

Further potential avenues of expansion overseas include a NICU in Gyula, Hungary, and Belgium.

14.2 Quantification of the Effect of Glycaemic Control on Clinical Outcomes

To date, the literature surrounding glycaemic control in premature infants has focused on clinical outcomes rather than first achieving control, and results are thus confounded by poor control and increased incidence of hypoglycaemia. As a glycaemic control tool, STAR-GRYPHON can be used to assess the long and short term effects of glycaemic control on clinical outcomes such as morbidity, length of hospital stay, and length of mechanical ventilation.

Long-term follow up of STAR-GRYPHON clinical results and outcomes, and comparison to retrospective data and outcomes, may allow retrospective examination of the effect of tight glycaemic control on clinical outcomes. While incomplete in itself to prove or disprove the benefits of glycaemic control, this analysis would add to the growing literature surrounding glycaemic control in the premature infant cohort.

STAR-GRYPHON is currently being used in the HINT2 randomised control trial to assess the affect of glycaemic control on outcomes. This, and other future trials, should enable more accurate and comprehensive analysis of the effect of glycaemic control on clinical outcomes.

14.3 Expansion of Approach into Paediatric Intensive Care

With model-based STAR glycaemic control protocols developed for neonatal and adult intensive care, the logical next step is to ‘close the gap’ and develop a model-based approach for use in paediatric intensive care.

Paediatric intensive care is another area where much time, effort, and resource has been spent in the last few years to determine and quantify the potential risks and benefits of glycaemic control. As in the neonatal intensive care setting, this work has been largely divorced from any real attempt to determine safe and effective protocols for dosing insulin, instead dealing with outcomes. There is thus scope for first the development of a glycaemic control tool, and then the use of it to more comprehensively and effectively determine the effect of glycaemic control on patient outcomes.

14.4 Quantification of External Contributions to Insulin Sensitivity

Within a clinical context, demands on clinical time and resources mean that a number of external factors can influence system performance by contributing to noise in the insulin sensitivity trace. Such factors can include, but are not limited to, absorption of insulin by IV tubing, dead space within the pump and IV line, and timing or data entry errors around nutrition and insulin doses or changes.

Insulin absorption by tubing has been reported in literature, with high rates of insulin absorption being reported even after pre-soaking of tubing in insulin. These studies tend to examine percentage absorption as a function of flow rate and/or insulin concentration. No study has comprehensively examined insulin absorption, accounting for surface area, concentration, and flow rate. Such a study can easily be carried out without risk or the need for clinical patients, and would allow more accurate modelling of insulin sensitivity during the first few hours of glycaemic control, thus improving control.

The glucose-insulin model relies on knowledge of system inputs. A sensitivity analysis can be carried out to examine the impact of timing errors and dead space within the IV tubing on insulin sensitivity and glycaemic control performance. Such an analysis would have twofold benefits. First, it would allow for a more comprehensive knowledge of the limitations of the system and the nature of model based insulin sensitivity. Second, it allows clinical practice around glycaemic control to be more informed; highlighting areas where clinical staff need to ensure accuracy of the information entered.

Many infusion pumps are now able to transmit information concerning rates and concentrations directly to electronic devices. At minimum, a one-way exchange of data with a model-based protocol on a tablet computer would allow the tablet computer to have more up to date and accurate information concerning system inputs, without increasing clinical workload. This would also reduce the effect of erroneous data entry on control, thereby improving the quality and effectiveness of glycaemic control.

14.5 Quantification of Physiological Influences on Insulin Sensitivity

Both the stress response and some types of medication are known to influence a patient's blood glucose and response to insulin. Observations on the correlations between patient condition and insulin sensitivity could potentially enable glycaemic control protocols to be

tailored to patient condition or medication regime. Further, it may also allow long term warning or diagnosis of changes in patient condition in cases such as pre-septic or septic infants.

14.6 Investigation of SGA and Sex Based Cohorts

Within this work both insulin secretion and insulin sensitivity were observed to differ between male and female infants. Analysis of insulin secretion over a larger and more frequently measured cohort would enable further investigation of sex based insulin secretion differences. In particular, quantitative analysis of insulin secretion in older infants and children would enable one aspect of the maturation of the glucose-insulin system to be examined, improving attempts at glycaemic control in these cohorts, and adding to current knowledge surrounding glycaemic development in children and young adults.

One result of this thesis was that small for gestational age (SGA) and male infants had higher insulin sensitivity than appropriate for gestational age (AGA) and female infants. This result was surprising, as these are two cohorts generally considered to be more vulnerable and with worsened outcomes. Long term follow up of clinical data from a number of different glycaemic control cohorts is required to confirm or negate these observations, and determine clinical implications for glycaemic control in these infants.

14.7 Validation of Gut Models

In Chapter 8 a two compartment gut model was developed for term and premature infant cohorts. This model requires validation for glycaemic control in virtual trials and clinical patients. Virtual trials will test the effect of the new model on glycaemic control outcomes, and, if required, enable protocols to be developed to cope with large peaks in model predictions due to glucose appearance from enteral feeds. Once virtual trials have assessed the likely performance and safety of the new model, it can be used in clinical trials to assess clinical efficacy and safety.

14.8 CGM use in Premature Infants

The low blood volume, small size, and inherent fragility of the very premature infant cohort means that clinical measurements must be minimised. Continuous glucose monitors (CGMs)

may provide a way by which the amount of glycaemic information can be increased, and clinical blood glucose measurements decreased.

Currently, CGMs are well known to suffer from noise and drift. While no CGM has been designed specifically for the term or premature infant cohort, several studies have used CGMs in these cohorts (Beardsall et al., 2005, Harris et al., 2013) to capture and quantify hypoglycaemic incidence.

Analysis of CGM data from premature infants will allow CGM models to be built and potentially enable CGMs to be integrated with STAR-GRYPHON. This would increase the amount of data available to the model, improving model-based insulin sensitivity, and providing pre-emptive warnings of downward trends in blood glucose during insulin therapy. Thus, the efficacy and safety of model-based glycaemic control protocols could be improved through the use of CGMs.

The End



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